U.S. Application No.: 10/625,056 Attorney Docket No. 6100-065-999

Reference No.: B24

(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization International Bureau





(43) International Publication Date 12 September 2002 (12.09.2002)

PCT

(10) International Publication Number WO 02/070682 A2

- (51) International Patent Classification⁷: C12N 13/00, 1/16, 1/04, C12P 1/02, A62D 3/00, B09B 3/00, C02F 3/00 // (C12P 1/02, C12R 1:645)
- (21) International Application Number: PCT/GB01/05439
- (22) International Filing Date:

11 December 2001 (11.12.2001)

(25) Filing Language:

English

(26) Publication Language:

English

(30) Priority Data:

09/797,377	1 March 2001 (01.03.2001)	US
09/797,382	1 March 2001 (01.03.2001)	US
09/797,378	1 March 2001 (01.03.2001)	US
09/797,371	1 March 2001 (01.03.2001)	US
09/797,381	1 March 2001 (01.03.2001)	US
09/797,372	1 March 2001 (01.03.2001)	US
09/797,493	1 March 2001 (01.03.2001)	US
09/797,437	1 March 2001 (01.03.2001)	US

- (71) Applicant (for all designated States except US): ULTRA BIOTECH LIMITED [GB/GB]; 60 Circular Road, 2nd Floor, Douglas, Isle of Man IM1 1SA (GB).
- (72) Inventor; and
- (75) Inventor/Applicant (for US only): CHEUNG, Ling, Yuk

[CN/CN]; Flat 11A, Tower 2, Deerhill Bay, Taipo, New Territories, Hong Kong (CN).

- (74) Agents: MCCALLUM, William, Potter et al.; Cruikshank & Fairweather, 19 Royal Exchange Square, Glasgow G1 3AE (GB).
- (81) Designated States (national): AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW.
- (84) Designated States (regional): ARIPO patent (GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Published:

 without international search report and to be republished upon receipt of that report

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

 \forall

(54) Title: METHODS AND COMPOSITIONS FOR WASTE TREATMENT

(57) A bstract: Compositions comprising one or more pluralities of yeast cells, wherein said pluralities of yeast cells characterized by a substantial increase in their capability to (1) to degrade polymeric compounds such as polysaccharides and plastics; (2) to degrade nitrogen-containing compounds such as proteins and nucleic acids; (3) to degrade environmental toxins, e.g., certain toxic chemicals such as organic solvents and antibiotics used in fodder and organic pesticides; (4) to convert biologically available nitrogen in a culture medium into their own biomass; (5) to convert biologically available phosphorus in a culture medium into their own biomass; (6) to reduce foul odor of malodorous materials; (7) to suppress the proliferation of pathogenic microorganisms; and/or (8) to suppress the gro with of algae or decompose algal debris as a result of having been cultured in the presence of an alternating electric field having a specific frequency and a specific field strength, as compared to yeast cells not having been so cultured. Also included are methods of making such compositions and methods of waste water treatment using these compositions.

WO 02/070682

BEST AVAILABLE COPY

WO02070682

Publication Title:

METHODS AND COMPOSITIONS FOR WASTE TREATMENT

Abstract:

Compositions comprising one or more pluralities of yeast cells, wherein said pluralities of yeast cells characterized by a substantial increase in their capability to (1) to degrade polymeric compounds such as polysaccharides and plastics; (2) to degrade nitrogen-containing compounds such as proteins and nucleic acids; (3) to degrade environmental toxins, e.g., certain toxic chemicals such as organic solvents and antibiotics used in fodder and organic pesticides; (4) to convert biologically available nitrogen in a culture medium into their own biomass; (5) to convert biologically available phosphorus in a culture medium into their own biomass; (6) to reduce foul odor of malodorous materials; (7) to suppress the proliferation of pathogenic microorganisms; and/or (8) to suppress the growth of algae or decompose algal debris as a result of having been cultured in the presence of an alternating electric field having a specific frequency and a specific field strength, as compared to yeast cells not having been so cultured. Also included are methods of making such compositions and methods of waste water treatment using these compositions.

Data supplied from the esp@cenet database - http://ep.espacenet.com

METHODS AND COMPOSITIONS FOR WASTE TREATMENT

CROSS REFERENCE TO OTHER APPLICATIONS

This application claims priority from United States Patent

5 Application Nos. 09/797,377, 09/797,382, 09/797,378, 09/797,371, 09/797,381, 09/797,372, 09/797,493, and 09/797,437, all of which filed March 1, 2001, the contents of which are incorporated herein by reference.

FIELD OF THE INVENTION

The invention relates to the use of yeast compositions in liquid
waste treatment. These yeast compositions can be obtained by growth in
electromagnetic fields with specific frequencies and field strengths.

BACKGROUND OF THE INVENTION

Environmental pollution by urban sewage and industrial waste water has posed a serious health threat to living organisms in the world. The main components of these water pollutants generally fall into eight categories: complex, high molecular weight, polymeric compounds, in particular, polysaccharides (e.g., starch or cellulose), lignin, polyethylene, polypropylene, polyvinyl chloride and polystyrene; complex, high molecular weight, nitrogen-containing compounds, such as proteins, polypeptides, amino acids, vitamins, lipids, and polynucleic acids; environmentally harmful toxins, e.g., antibiotics, undesirable chemicals, such as phenols, sulfur-containing compounds, alkanes, and tricholomethane, and chemical

additives commonly found in fertilizers and detergents; water-soluble nitrogen compounds, such as ammonium salts, nitrates, nitrites, and amino acids; biologically available phosphates, such as HPO₄²⁻ and H₂PO₄; odorous materials, such as ammonia, sulfides and fatty acids; pathogenic microorganisms, such as *E. Coli, Salmonella*, bacteria, fungi, actinomyces, and different viruses; and algae, such as green algae, blue algae, and red algae. Currently, the most common methods for large-scale water treatment include the activated sludge technology and the biomembrane technology. These technologies rely on the innate abilities of myriad natural microorganisms, such as fungi, bacteria and protozoa, to degrade pollutants. However, the compositions of these natural microbial components are difficult to control, affecting the reproducibility and quality of water treatment. Moreover, pathogenic microbes existing in these activated sludge or biomembranes cannot be selectively inhibited, and such microbes usually enter the environment with the treated water, causing "secondary pollution."

Further, most of the current technologies cannot degrade harmful chemicals such as pesticides, insecticides, and chemical fertilizers. These technologies also cannot alleviate eutrophication, another serious environmental problem around the world. Eutrophication is usually caused by sewage, industrial waste water, fertilizers and the like. It refers to waters (e.g., a lake or pond) rich in mineral and organic nutrients that promote a proliferation of plant life, especially algae, which reduces the dissolved oxygen content or otherwise deteriorates water quality. Eutrophication often results in the extinction of other organisms.

15

20

25

30

SUMMARY OF THE INVENTION

This invention is based on the discovery that certain yeast cells can be activated by electromagnetic fields having specific frequencies and field strengths to degrade or convert environmental pollutants to harmless final products. Compositions comprising these activated yeast cells can therefore be useful for waste treatment, for example, treatment of sewage, industrial waste water, surface water, drinking water, sediment, soil, garbage, and manure. Waste treatment methods using the compositions are more effective, efficient and economical than conventional methods.

This invention embraces a composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 4230 to 4260 MHz (e.g., 4240-4260 MHz), and a field strength in the range of about 0.5 to 360 mV/cm (e.g., 80-320, 80-300, 60-270, 90-320, 70-350, or 60-260 mV/cm). The yeast cells are cultured in the alternating electric field for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to degrade polymeric compounds in a culture medium. An exemplary period of time is about 12 to 400 hours, e.g., 220-360, 190-370, 160-280, 140-280, 222-382, 220-380, or 230-380 hours. Yeast species 10 useful in this composition include, but are not limited to, Saccharomyces cerevisiae, Saccharomyces carlsbergensis, and Hansenula subpelliculosa. For instance, the yeast cells can be of the strain Saccharomyces cerevisiae Hansen AS2.11, AS2.53, AS2.56, AS2.70, AS2.98, AS2.101, AS2.168, AS2.374, AS2.406, AS2.409, AS2.430, AS2.453, AS2.463, AS2.467, AS2.502, AS2.516, 15 AS2.536, AS2.541, or IFFI1331; Saccharomyces carlsbergensis AS2.443 or AS2.459; or Hansenula subpelliculosa Bedford AS2.738 or AS2.740.

This invention embraces a composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 5520 to 5540 MHz (e.g., 5521-5538 MHz) and a field strength in the range of about 0.5 to 360 mV/cm (e.g., 90-360 or 120-340 mV/cm). The yeast cells are cultured in the alternating electric field for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to degrade nitrogen-containing compounds in a culture medium. An exemplary period of time is about 12-450 hours, e.g., 228-424 or 208-320 hours. Yeast species useful in this composition include, but are not limited to, *Saccharomyces cerevisiae* and *Saccharomyces carlsbergensis*. For instance, the yeast cells can be of the strain *Saccharomyces cerevisiae* Hansen AS2.93, AS2.98, AS2.152, AS2.423, AS2.452, AS2.458, AS2.502, AS2.535, or AS2.561; or *Saccharomyces carlsbergensis* AS2.440 or AS2.595.

20

Embraced within this invention is also a composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 70 to 100 MHz (e.g., 70, 71, 72, 73, 74, 75, 76,

77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, or 100 MHz) and a field strength in the range of about 0.5 to 350 mV/cm (e.g., 30-220, 30-230, 30-250, 90-280, 80-280, 100-200, 110-280, 100-220, 116-225, 120-280, 90-190, 100-190, 160-300, 120-300, 200-300, or 130-310 mV/cm). The yeast cells are cultured in the alternating electric field for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to degrade environmental toxins, such as antibiotics and organic solvents, in a culture medium. An exemplary period of time is about 12-400 hours, e.g., 180-328, 114-244, 80-380, 80-365, 120-350, 90-330, 130-330, 100-280, 110-330, 130-290, 80-290, 110-360, or 110-340 hours.

This invention also embraces a composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 126 to 142 MHz (e.g., 126, 127, 128, 129, 130, 131, 132, 133, 134, 135, 136, 137, 138, 139, 140, 141, or 142 MHz) and a field strength in the range of about 0.5 to 350 mV/cm (e.g., 90-280 mV/cm) for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to degrade trichloromethane in a culture medium.

10

20

This invention also embraces a composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 52 to 70 MHz (e.g., 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, or 70 MHz) and a field strength in the range of about 0.5 to 350 mV/cm (e.g., 80-280 mV/cm) for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to degrade toluene or ethylbenzene in a culture medium.

This invention also embraces a composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 30 to 50 MHz (e.g., 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, or 50 MHz) and a field strength in the range of about 0.5 to 350 mV/cm (e.g., 80-280 mV/cm) for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to degrade p-xylene in a culture medium.

This invention embraces yet another composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 200 to 220 MHz (e.g., 200, 201, 202, 203, 204, 205, 206, 207, 208, 209, 210, 211, 212, 213, 214, 215, 216, 217, 218, 219, or 220 MHz) and a field strength in the range of about 0.5 to 350 mV/cm (e.g., 80-280 mV/cm) for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to degrade furazolidonum in a culture medium.

This invention embraces another composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 213 to 229 MHz (e.g., 213, 214, 215, 216, 217, 218, 219, 220, 221, 222, 223, 224, 225, 226, 227, 228, or 229 MHz) and a field strength in the range of about 0.5 to 350 mV/cm (e.g., 90-280 mV/cm) for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to degrade decoquinate in a culture medium.

This invention also embraces a composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 133 to 151 MHz (e.g., 133, 134, 135, 136, 137, 138, 139, 140, 141, 142, 143, 144, 145, 146, 147, 148, 149, 150, or 151 MHz) and a field strength in the range of about 0.5 to 350 mV/cm (e.g., 120-280 mV/cm) for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to degrade benzaldehyde in a culture medium.

15

20

This invention also embraces a composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 145 to 162 MHz (e.g., 145, 146, 147, 148, 149, 150, 151, 152, 153, 154, 155, 156, 157, 158, 159, 160, 161, or 162 MHz) and a field strength in the range of about 0.5 to 350 mV/cm (e.g., 100-200 mV/cm) for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to degrade propylaldehyde in a culture medium.

This invention also embraces a composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 156 to 176 MHz (e.g., 156, 157, 158, 159, 160, 161, 162, 163, 164, 165, 166, 167, 168, 169, 170, 171, 172, 173, 174, 175, or 176

MHz) and a field strength in the range of about 0.5 to 350 mV/cm (e.g., 110-280 mV/cm) for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to degrade enanthaldehyde in a culture medium.

This invention also embraces a composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 163 to 183 MHz (e.g., 163, 164, 165, 166, 167, 168, 169, 170, 171, 172, 173, 174, 175, 176, 177, 178, 179, 180, 181, 182, or 183 MHz) and a field strength in the range of about 0.5 to 350 mV/cm (e.g., 100-220 mV/cm) for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to degrade m-dichlorobenzene in a culture medium.

This invention embraces yet another composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 175 to 191 MHz (e.g., 175, 176, 177, 178, 179, 180, 181, 182, 183, 184, 185, 186, 187, 188, 189, 190, or 191 MHz) and a field strength in the range of about 0.5 to 350 mV/cm (e.g., 116-225 mV/cm) for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to degrade acetophenone in a culture medium.

This invention also embraces a composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 183 to 205 MHz (e.g., 183, 184, 185, 186, 187, 188, 189, 190, 191, 192, 193, 194, 195, 196, 197, 198, 199, 200, 201, 202, 203, 204, or 205 MHz) and a field strength in the range of about 0.5 to 350 mV/cm (e.g., 90-190 mV/cm) for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to degrade arsanilic acid in a culture medium.

20

25

30

This invention also embraces a composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 114 to 128 MHz (e.g., 114, 115, 116, 117, 118, 119, 120, 121, 122, 123, 124, 125, 126, 127, or 128 MHz) and a field strength in the range of about 0.5 to 350 mV/cm (e.g., 100-190 mV/cm) for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to degrade roxarsone in a culture medium.

This invention embraces another composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 244 to 264 MHz (e.g., 244, 245, 246, 247, 248, 249, 250, 251, 252, 253, 254, 255, 256, 257, 258, 259, 260, 261, 262, 263, or 264 MHz) and a field strength in the range of about 0.5 to 350 mV/cm (e.g., 160-300 mV/cm) for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to degrade dodecane in a culture medium.

This invention also embraces a composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 252 to 278 MHz (e.g., 252, 253, 254, 255, 256, 257, 258, 259, 260, 261, 262, 263, 264, 265, 266, 267, 268, 269, 270, 271, 272, 273, 274, 275, 276, 277, or 278 MHz) and a field strength in the range of about 0.5 to 350 mV/cm (e.g., 120-300 mV/cm) for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to degrade nonadecane or octacosane in a culture medium.

10

15

20

25

30

This invention also embraces a composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 220 to 250 MHz (e.g., 220, 222, 224, 226, 228, 230, 232, 234, 236, 238, 240, 242, 244, 246, 248, or 250 MHz) and a field strength in the range of about 0.5 to 350 mV/cm (e.g., 200-300 mV/cm) for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to degrade trichlorphonum in a culture medium.

This invention embraces yet another composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 220 to 250 MHz (e.g., 220, 222, 224, 226, 228, 230, 232, 234, 236, 238, 240, 242, 244, 246, 248, or 250 MHz) and a field strength in the range of about 0.5 to 350 mV/cm (e.g., 130-310 mV/cm) for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to degrade dinitomidum or zoalene in a culture medium.

Yeast species useful in the above seventeen compositions include, but are not limited to, Saccharomyces cerevisiae, Saccharomyces carlsbergensis, Saccharomyces rouxii, and Candida utilis. For instance, the yeast cells can be of

the strain Saccharomyces cerevisiae AS2.4, AS2.14, AS2.416, AS2.430, AS2.593, IFFI1002, IFFI1006, IFFI1043, IFFI1045, IFFI1048, IFFI1063, IFFI1059, IFFI1206, IFFI1209, IFFI1210, IFFI1211, IFFI1213, IFFI1215, IFFI1220, IFFI1221, IFFI1224, IFFI1248, IFFI1270, IFFI1290, IFFI1291, IFFI1293, IFFI1297, IFFI1301, IFFI1302, IFFI1310, IFFI1311, IFFI1331, IFFI1335, IFFI1336, IFFI1338, IFFI1339, IFFI1340, IFFI1345, IFFI1396, IFFI1399, IFFI1411, or IFFI1413; Saccharomyces willianus Saccardo AS2.293; Saccharomyces carlsbergensis AS2.377 or AS2.444; Saccharomyces rouxii AS2.178; or Candida utilis AS2.120.

This invention further includes a composition comprising a plurality 10 of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 660 to 680 MHz (e.g., 660, 661, 662, 663, 664, 665, 666, 667, 668, 669, 670, 671, 672, 673, 674, 675, 676, 677, 678, 679, or 680 MHz) and a field strength in the range of about 0.1 to 350 mV/cm (e.g., 140-320 or 120-290 mV/cm). The yeast cells are cultured for a period of time sufficient to 15 substantially increase the capability of said plurality of yeast cells to convert biologically available nitrogen in a culture medium into intracellular nitrogen. An exemplary period of time is about 12-420 hours (e.g., 192-304 or 226-412 hours). This invention also includes a composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of 20 about 2160 to 2190 MHz (e.g, 2170 to 2185 MHz) and a field strength in the range of about 0.1 to 350 mV/cm (e.g., 140-320 mV/cm) for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to convert ammonium in a culture medium into intracellular nitrogen. Yeast species useful in these compositions include, but are not limited to, Saccharomyces cerevisiae, 25 Candida tropicalis and Geotrichum candidum. For instance, the yeast cells can be of the strain Saccharomyces cerevisiae AS2.196, AS2.336, AS2.400, AS2.416, AS2.423, or AS2.982; Saccharomyces willianus Saccardo AS2.152 or AS2.614; Candida tropicalis AS2.1387; or Geotrichum candidum AS2.498.

This invention also embraces a composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 80 MHz to 440 MHz (e.g., 86-120 or 410-430

30

MHz) and a field strength in the range of about 0.5 to 350 mV (e.g., 60-260 mV/cm). The yeast cells are cultured for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to convert biologically available phosphorus in a culture medium into intracellular phosphorus. An exemplary period of time is about 12-400 hours, e.g., 228-368 hours. Yeast species useful in this composition include, but are not limited to, Saccharomyces cerevisiae and Saccharomyces carlsbergensis. For instance, the yeast cells can be of the strain Saccharomyces cerevisiae AS2.346, AS2.423, AS2.430, AS2.451, AS2.558, AS2.620, AS2.628, or IFFI1203; or Saccharomyces carlsbergensis AS2.189.

10

20

25

This invention embraces a composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 2160 to 2380 MHz (e.g., 2160-2250 or 2280-2380 MHz) and a field strength in the range of about 0.5 to 320 mV/cm (e.g., 40-260, 70-260, 80-250, 90-260, or 140-300 mV/cm). The yeast cells are cultured for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to reduce odor in malodorous materials. An exemplary period of time is about 12 to 350 hours (e.g., 70-220, 70-320, 80-310, 85-220, 110-230, or 120-300 hours). Yeast species useful in this composition include, but are not limited to, *Saccharomyces cerevisiae* and *Saccharomyces carlsbergensis*. For instance, the yeast cells can be of the strain *Saccharomyces cerevisiae* Hansen AS2.53, AS2.163, AS2.396, AS2.397, AS2.423, AS2.452, AS2.502, AS2.516, AS2.541, AS2.558, AS2.559, AS2.560, AS2.561, AS2.561, AS2.562, AS2.607, AS2.612, IFFI 1052, IFFI 1202, IFFI 1213, IFFI 1247, or IFFI 1397; or *Saccharomyces carlsbergensis* Hansen AS2.605.

Embraced within this invention is also a composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 30 to 50 MHz (e.g., 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, or 50 MHz) and a field strength in the range of about 0.5 to 200 mV/cm (e.g., about 10 to 180 mV/cm). The yeast cells are cultured in the presence of the alternating electric field for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to suppress the proliferation of pathogenic microorganisms. An exemplary period of

time is about 12-300 hours (e.g., 144-272 hours). Yeast species useful in this composition include, but are not limited to, Saccharomyces cerevisiae, Saccharomyces carlsbergensis, Saccharomyces uvarum, and Saccharomyces willianus. For instance, the yeast cells can be of the strain Saccharomyces cerevisiae Hansen ACCC2034, ACCC2043, AS2.70, AS2.369, AS2.408, AS2.451, AS2.562, AS2.607, IFFI1021, IFFI1037, IFFI1211, IFFI1221, IFFI1251, IFFI1301, IFFI1307, IFFI1308, IFFI1331, or IFFI1345; Saccharomyces carlsbergensis Hansen AS2.200; Saccharomyces uvarum Beijer IFFI1023, IFFI1032, or IFFI1205; or Saccharomyces willianus Saccardo AS2.119 or AS2.152.

10

15

20

25

30

Embraced within this invention is also a composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 6340 to 6380 MHz (e.g., 6352-6370 MHz) and a field strength in the range of about 0.5 to 400 mV/cm (e.g., 70-310, 100-330, or 120-360 mV/cm). The yeast cells are cultured for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to suppress the growth of algae. An exemplary period of time is about 12-450 hours (e.g., 256-432 hours). This invention also embraces a composition comprising a plurality of yeast cells that have been cultured in an alternating electric field having a frequency in the range of about 4440 to 4470 MHz (e.g., 4452-4470 MHz) and a field strength in the range of about 0.5 to 400 mV/cm (e.g., 50-280 mV/cm). The yeast cells are cultured for a period of time sufficient to substantially increase the capability of said plurality of yeast cells to decompose algae. An exemplary period of time is about 12-600 hours (e.g., 320-576 hours). Yeast species useful in these compositions include, but are not limited to, Saccharomyces cerevisiae. For instance, the yeast cells can be of the strain Saccharomyces cerevisiae Hansen AS2.408, AS2.414, AS2.416, AS2.422, AS2.453, AS2.486, AS2.558, AS2.562, or IFFI1292.

In one embodiment, the frequency and/or the field strength of the alternating electric field can be altered within the aforementioned ranges during said periods of time. In other words, the yeast cells can be exposed to a series of electromagnetic fields.

Yeast cells that can be included in this invention are available from the China General Microbiological Culture Collection Center ("CGMCC"), a depository recognized under the Budapest Treaty (China Committee for Culture Collection of Microorganisms, Institute of Microbiology, Chinese Academy of Sciences, Haidian, P.O. Box 2714, Beijing, 100080, China).

This invention further embraces a composition comprising a plurality of yeast cells, wherein said plurality of yeast cells have been activated such that they have a substantially increased capability (1) to degrade polymeric compounds, (2) to degrade nitrogen-containing compounds, (3) to degrade environmental toxins, such as antibiotics and other organic compounds of interests, (4) to convert biologically available nitrogen in a culture medium into intracellular nitrogen, (5) to convert biologically available phosphorus in a culture medium into intracellular phosphorus, (6) to reduce odor in malodorous materials, (7) to suppress the proliferation of pathogenic microorganisms, or (8) to suppress the growth of algae or decompose algal debris, as compared to unactivated yeast cells. Included in this invention are also methods of making these compositions. A composition comprising two or more of the above-described compositions is also within the scope of this invention.

10

15

20

The compositions described herein can be used for waste water treatment. Two or more of these compositions can also be mixed in accordance with the types of pollutants in the waste water to achieve optimal water treatment results. For convenience, each of the compositions in such a mixture is called herein a "yeast component:"

As used herein, a "substantially increase" means an increase of more than 10 (e.g., 10^2 , 10^3 , 10^4 , 10^5 , or 10^6) fold.

A "culture medium" refers to a medium used in a laboratory for selecting and growing a given yeast strain, or to liquid or solid waste in need of treatment.

"Polymeric compounds" refer to high molecular weight organic

compounds consisting of repeated, linked subunits. Exemplary polymeric compounds include, but are not limited to, polysaccharides (e.g., starch or cellulose), lignin, polyethylene, polypropylene, polyvinyl chloride and polystyrene.

"Nitrogen-containing compounds" refer to complex, high molecular weight, nitrogen-containing compounds, including but not limited to proteins, peptides, lipids, and nucleic acids.

Antibiotics degradable by the yeast compositions of the invention

5 include, but are not limited to, beta-lactams, tetracyclines, polypeptides,
glycopeptides, aminoglycosides, and macrolides. Specific examples of antibiotics
are penicillin, aureomycin, chlortetracycline, oxytetracycline, doxycycline,
tetracycline, streptomycin, kanamycin, erythromycin, spiramycin, and bacitracin.
Organic solvents degradable by the yeast compositions of this invention include,
but are not limited to, trichloromethane, toluene, ethylbenzene, p-xylene,
furazolidonum, decoquinate, benzaldehyde, propylaldehyde, enanthaldehyde, mdichlorobenzene, acetophenone, arsanilic acid, roxarsone, dodecane, nonadecane,
octacosane, trichlorophonum, dinitomidum and zoalene.

"Biologically available," "bio-available," "biologically assimilable," or "bio-assimilable" nitrogen refers to nitrogen that is readily available, useable, or assimilable by living organisms for survival and/or growth. Exemplary bio-available or bio-assimilable nitrogen includes, but is not limited to, NH₄⁺, NO₃⁻ and NO₂⁻, other water-soluble inorganic nitrogen-containing compounds, and organic nitrogen-containing compounds.

15

20

25

30

"Biologically available" or "biologically assimilable" phosphorus refers to phosphorus that is readily available, useable, or assimilable by living organisms for survival and/or growth. Exemplary biologically available or assimilable phosphorus includes, but is not limited to, PO₄³⁻, HPO₄²⁻, H₂PO₄⁻, other water-soluble inorganic phosphorus-containing compounds, and organic phosphorus-containing compounds.

"Reducing odor" or "deodorizing" refers to a process which results in a lower concentration of one or more odorous compounds. Odorous compounds include, but are not limited to, hydrogen sulfide, ammonium sulfide, other sulfurcontaining compounds, ammonia, indole, methylindoles, p-cresol, amines such as methylamine, dimethylamine and trimethylamine, and odorous organic acids, such as carboxylic acids, e.g., formic acid, acetic acid, propanoic acid and butyric acid, and other volatile fatty acids.

"Suppressing the growth of pathogenic microbes" means preventing the increase in, or even decreasing, the number of pathogenic microorganisms. It is to be understood that in the absence of yeast cells of this invention, the number of pathogenic microbes will increase naturally over a period of time. Pathogenic microorganisms include, but are not limited to, bacteria such as those belonging to the Escherichia, Salmonella, Shigella, Mycobacterium, Staphylococcus, Bacillus, Streptococcus and Diplococcus genera.

"Suppressing the growth of algae" means preventing the increase in or even reducing the proliferation rate of algae. "Decomposing algae" means breaking down debris of algae into harmless products. It is to be understood that in the absence of yeast cells of this invention, the number of algae will increase naturally over a period of time. Algae include, but are not limited to, green, blue, and red algae.

Unless otherwise defined, all technical and scientific terms used

herein have the same meaning as commonly understood by one of ordinary skill in
the art to which this invention belongs. Exemplary methods and materials are
described below, although methods and materials similar or equivalent to those
described herein can also be used in the practice or testing of the present invention.
All publications and other references mentioned herein are incorporated by
reference in their entirety. In case of conflict, the present specification, including
definitions, will control. The materials, methods, and examples are illustrative
only and not intended to be limiting.

Other features and advantages of the invention will be apparent from the following detailed description, and from the claims.

BRIEF DESCRIPTION OF THE DRAWINGS

25

30

Fig. 1 is a schematic diagram showing an exemplary apparatus for activating yeast cells using electromagnetic fields. 1: yeast culture; 2: container; 3: power supply.

DETAILED DESCRIPTION OF THE INVENTION

This invention is based on the discovery that certain yeast strains can be activated by electromagnetic fields ("EMF") having specific frequencies and field strengths to become highly efficient in (1) degrading polymeric compounds such as polysaccharides and plastics, (2) degrading nitrogen-containing compounds 5 such as proteins and nucleic acids, (3) degrading environmental toxins, e.g., certain toxic chemicals such as organic solvents and antibiotics used in fodder and organic pesticides, (4) converting biologically available nitrogen in a culture medium into intracellular nitrogen (i.e., the yeast cells incorporate biologically available nitrogen in their environs into their own biomass), (5) converting biologically available 10 phosphorus in a culture medium into intracellular phosphorus (i.e., the yeast cells incorporate biologically available phosphorus in their environs into their own biomass), (6) reducing foul odor of malodorous materials, (7) suppressing the proliferation of pathogenic microorganisms, and/or (8) suppressing the growth of harmful algae or decomposing algal debris. As used herein, yeast cells with one of the above eight functions are defined herein as belonging to the same "function" or "functional group." Compositions containing these activated yeast cells, either of the same function or of two or more different functions, are useful in waste treatment, in particular, waste water treatment.

Without being bound by any theory or mechanism, the inventor believes that EMFs activate or enhance the expression of a gene or a set of genes in yeast cells such that the yeast cells become active or more efficient in performing certain metabolic activities which lead to the respective, desired water treatment functions.

I. Yeast Strains Useful in the Invention

20

The types of yeasts useful in this invention include, but are not limited to, yeasts of the genera of Saccharomyces, Schizosaccharomyces, Sporobolomyces, Torulopsis, Trichosporon, Wickerhamia, Ashbya, Blastomyces, Candida, Citeromyces, Crebrothecium, Cryptococcus, Debaryomyces, Endomycopsis, Eremothecium, Geotrichum, Hansenula, Kloeckera, Lipomyces, Pichia, Rhodosporidium, and Rhodotorula.

Exemplary species within the above-listed genera include, but are not limited to, Saccharomyces cerevisiae, Saccharomyces bailii, Saccharomyces

carlsbergensis, Saccharomyces chevalieri, Saccharomyces delbrueckii, Saccharomyces exiguus, Saccharomyces fermentati, Saccharomyces logos, Saccharomyces mellis, Saccharomyces microellipsoides, Saccharomyces oviformis, Saccharomyces rosei, Saccharomyces rouxii, Saccharomyces sake, Saccharomyces 5 uvarum, Saccharomyces willianus, Saccharomyces sp., Saccharomyces ludwigii, Saccharomyces sinenses, Saccharomyces bailii, Saccharomyces carlsbergensis, Schizosaccharomyces octosporus, Schizosaccharomyces pombe, Sporobolomyces roseus, Sporobolomyces salmonicolor, Torulopsis candida, Torulopsis famta, Torulopsis globosa, Torulopsis inconspicua, Trichosporon behrendoo, 10 Trichosporon capitatum, Trichosporon cutaneum, Wickerhamia fluoresens, Ashbya gossypii, Blastomyces dermatitidis, Candida albicans, Candida arborea, Candida guilliermondii, Candida krusei, Candida lambica, Candida lipolytica, Candida parakrusei, Candida parapsilosis, Candida pseudotropicalis, Candida pulcherrima, Candida robusta, Candida rugousa, Candida tropicalis, Candida 15 utilis, Citeromyces matritensis, Crebrothecium ashbyii, Cryptococcus laurentii, Cryptococcus neoformans, Debaryomyces hansenii, Debaryomyces kloeckeri, Debaryomyces sp., Endomycopsis fibuligera, Eremothecium ashbyii, Geotrichum candidum, Geotrichum ludwigii, Geotrichum robustum, Geotrichum suaveolens, Hansenula anomala, Hansenula arabitolgens, Hansenula jadinii, Hansenula 20 saturnus, Hansenula schneggii, Hansenula subpelliculosa, Kloeckera apiculata, Lipomyces starkeyi, Pichia farinosa, Pichia membranaefaciens, Rhodosporidium toruloides, Rhodotorula aurantiaca, Rhodotorula glutinis, Rhodotorula minuta,

Yeast strains useful in this invention can be obtained from
laboratory cultures, or from publically accessible culture depositories, such as
CGMCC and the American Type Culture Collection, 10801 University Boulevard,
Manassas, VA 20110-2209.

Rhodotorula rubar, and Rhodotorula sinesis.

For degradation of polymeric compounds such as polysaccharides and plastics, non-limiting examples of useful strains (with the accession numbers of CGMCC) are *Saccharomyces cerevisiae* Hansen AS2.11, AS2.53, AS2.56, AS2.70, AS2.98, AS2.101, AS2.168, AS2.374, AS2.406, AS2.409, AS2.430, AS2.453, AS2.463, AS2.467, AS2.502, AS2.516, AS2.536, AS2.541, and

IFFI1331; Saccharomyces carlsbergensis AS2.443 and AS2.459; and Hansenula subpelliculosa Bedford AS2.738 and AS2.740.

For degradation of nitrogen-containing compounds such as proteins and nucleic acids, non-limiting examples of yeast strains useful in this invention are *Saccharomyces cerevisiae* Hansen AS2.93, AS2.98, AS2.152, AS2.423, AS2.452, AS2.458, AS2.502, AS2.535, and AS2.561; and *Saccharomyces carlsbergensis* AS2.440 and AS2.595.

For degradation of environmental toxins, e.g., certain toxic chemicals such as organic solvents, antibiotics, pesticides and insecticides, non-limiting examples of useful strains are Saccharomyces cerevisiae AS2.4, AS2.14, AS2.416, AS2.430, AS2.593, IFFI1002, IFFI1006, IFFI1043, IFFI1045, IFFI1048, IFFI1063, IFFI1059, IFFI1206, IFFI1209, IFFI1210, IFFI1211, IFFI1213, IFFI1215, IFFI1220, IFFI1221, IFFI1224, IFFI1248, IFFI1270, IFFI1290, IFFI1291, IFFI1293, IFFI1297, IFFI1301, IFFI1302, IFFI1310, IFFI1311, IFFI1331, IFFI1335, IFFI1336, IFFI1338, IFFI1339, IFFI1340, IFFI1345, IFFI1396, IFFI1399, IFFI1411, and IFFI1413; Saccharomyces willianus Saccardo AS2.293; Saccharomyces carlsbergensis AS2.377 and AS2.444; Saccharomyces rouxii AS2.178; and Candida utilis AS2.120.

For conversion of biologically available nitrogen in a culture
medium into intracellular nitrogen, non-limiting examples of useful strains are
Saccharomyces cerevisiae AS2.196, AS2.336, AS2.400, AS2.416, AS2.423 and
AS2.982; Saccharomyces willianus Saccardo AS2.152 and AS2.614; Candida
tropicalis AS2.1387; and Geotrichum candidum AS2.498.

For conversion of biologically available phosphorus in a culture
medium into intracellular phosphorus, non-limiting examples of useful strains are
Saccharomyces cerevisiae Hansen AS2.346, AS2.423, AS2.430, AS2.451,
AS2.558, AS2.620, AS2.628, and IFFI1203; and Saccharomyces carlsbergensis
AS2.189.

For elimination of foul odor in malodorous materials, non-limiting examples of useful strains are *Saccharomyces cerevisiae* Hansen AS2.53, AS2.163, AS2.396, AS2.397, AS2.423, AS2.452, AS2.502, AS2.516, AS2.541, AS2.558, AS2.559, AS2.560, AS2.561, AS2.562, AS2.607, AS2.612, IFFI 1052, IFFI 1202,

IFFI 1213, IFFI 1247, and IFFI 1397; and Saccharomyces carlsbergensis Hansen AS2.605.

For suppression of the proliferation of pathogenic microorganisms, non-limiting examples of useful strains are *Saccharomyces cerevisiae* Hansen ACCC2034, ACCC2043, AS2.70, AS2.369, AS2.408, AS2.451, AS2.562, AS2.607, IFFI1021, IFFI1037, IFFI1211, IFFI1221, IFFI1251, IFFI1301, IFFI1307, IFFI1308, IFFI1331, and IFFI1345; *Saccharomyces carlsbergensis* Hansen AS2.200; *Saccharomyces uvarum* Beijer IFFI1023, IFFI1032, and IFFI1205; and *Saccharomyces willianus* Saccardo AS2.119 and AS2.152.

For suppression of the growth of harmful algae or decomposition of algal debris, non-limiting examples of useful strains are *Saccharomyces cerevisiae* Hansen AS2.408, AS2.414, AS2.416, AS2.422, AS2.453, AS2.486, AS2.558, AS2.562, and IFFI1292.

Although it is preferred, the preparation of the yeast compositions of
this invention is not limited to starting with a pure strain of yeast for each water
treatment function. A yeast composition of a particular water treatment function
may be produced by culturing a mixture of yeast cells belonging to different species
or strains that have the same water treatment function. The ability of any species or
strain of yeast to perform these desired functions can be readily tested by methods
known in the art. See also discussions below.

Certain yeast species that can be activated according to the present invention are known to be pathogenic to human and/or other living organisms. These yeast species include, for example, Ashbya gossypii, Blastomyces dermatitidis, Candida albicans, Candida parakrusei, Candida tropicalis,

Citeromyces matritensis, Crebrothecium ashbyii, Cryptococcus laurentii,

Cryptococcus neoformans, Debaryomyces hansenii, Debaryomyces kloeckeri,

Debaryomyces sp., and Endomycopsis fibuligera. Under certain circumstances, it may be less preferable to use such pathogenic yeasts in this invention. If use of these species is necessary, caution should be exercised to minimize the leak of the yeast cells into the final treatment product that enters the environment.

II. Application of Electromagnetic Fields

10

An electromagnetic field useful in this invention can be generated and applied by various means well known in the art. For instance, the EMF can be generated by applying an alternating electric field or an oscillating magnetic field.

Alternating electric fields can be applied to cell cultures through

electrodes in direct contact with the culture medium, or through electromagnetic induction. See, e.g., Fig. 1. Relatively high electric fields in the medium can be generated using a method in which the electrodes are in contact with the medium.

Care must be taken to prevent electrolysis at the electrodes from introducing undesired ions into the culture and to prevent contact resistance, bubbles, or other features of electrolysis from dropping the field level below that intended.

Electrodes should be matched to their environment, for example, using Ag-AgCl electrodes in solutions rich in chloride ions, and run at as low a voltage as possible. For general review, see Goodman et al., Effects of EMF on Molecules and Cells, International Review of Cytology, A Survey of Cell Biology, Vol. 158, Academic Press, 1995.

The EMFs useful in this invention can also be generated by applying an oscillating magnetic field. An oscillating magnetic field can be generated by oscillating electric currents going through Helmholtz coils. Such a magnetic field in turn induces an electric field.

The frequencies of EMFs useful in this invention range from 5 MHz to 10000 MHz. The field strength of the electric field useful in this invention ranges from about 0.1 to 400 mV/cm. The preferred frequency and field strength ranges for each water treatment function are described in detail below.

20

25

30

When a series of EMFs are applied to a yeast culture, the yeast culture can remain in the same container while the same set of EMF generator and emitters is used to change the frequency and/or field strength. The EMFs in the series can each have a different frequency or a different field strength; or a different frequency and a different field strength. Such frequencies and field strengths are preferably within the above-described ranges. In one embodiment, an EMF at the beginning of the series has a field strength identical to or lower than that of a subsequent EMF, such that the yeast cell culture is exposed to EMFs of progressively increasing field strength. Although any practical number of EMFs

can be used in a series, it may be preferred that the yeast culture be exposed to a total of 2, 3, 4, 5, 6, 7, 8, 9 or 10 EMFs in a series.

Fig. 1 illustrates an exemplary apparatus for generating alternating electric fields. An electric field of a desired frequency and intensity is generated by an AC source (3) capable of generating an alternating electric field, preferably in a sinusoidal wave form, in the frequency range of 5 to 10,000 MHz. Signal generators capable of generating signals with a narrower frequency range can also be used. If desirable, a signal amplifier can also be used to increase the output. The alternating electric field can be applied to the culture by a variety of means including placing the yeast culture in close proximity to the signal emitters. In one embodiment, the electric field is applied by electrodes submerged in the culture (1). In this embodiment, one of the electrodes can be a metal plate placed on the bottom of the container (2), and the other electrode can comprise a plurality of electrode wires evenly distributed in the culture (1) so as to achieve even distribution of the electric field energy. The number of electrode wires used depends on the volume of the culture as well as the diameter of the wires. In a preferred embodiment, for a culture having a volume up to 5000 ml, one electrode wire having a diameter of 0.1 to 1.2 mm can be used for each 100 ml of culture. For a culture having a volume greater than 1000 L, one electrode wire having a diameter of 3 to 30 mm can be used for each 1000 L of culture.

1. <u>Degradation of Polymeric Compounds</u>

20

30

The frequencies of EMFs useful for degrading polymeric compounds range from 5 MHz to 5000 MHz, e.g., from 4230 MHz to 4260 MHz. Exemplary frequencies are 4240, 4241, 4242, 4243, 4244, 4245, 4246, 4247, 4248, 4249, 4250, 4251, 4252, 4253, 4254, 4255, 4256, 4257, 4258, 4259 and 4260 MHz. The field strength of the electric field useful for this water treatment function ranges from about 0.5 mV/cm to 360 mV/cm, for example, from about 50 to 360 mV/cm (e.g., 80-320, 80-300, 60-270, 90-320, 70-350, or 60-260 mV/cm). Exemplary field strengths are 78, 82, 87, 90, 95, 108, 110, 240, 245, 250, 280, and 300 mV/cm.

By way of example, the yeast cells can be cultured in a first series of alternating electric fields each having a frequency in the range of 4240 to 4260

MHz and a field strength in the range of 50 to 360 mV/cm. The yeast cells are exposed to each EMF for about 10 to 30 hours. After the first series of culturing, the resultant yeast cells are further incubated in a second series of alternating electric fields for a total of 20 to 140 hours. It may be preferred that the frequencies in the second series of alternating electric fields are identical to those of the first series in sequence and the field strengths in the second series are increased to a higher level within the range of 50 to 360 mV/cm.

Although the yeast cells can be activated after even a few hours of culturing in the presence of an EMF, it may be preferred that the activated yeast cells be allowed to multiply and grow in the presence of the EMF(s) for a total of 220-360, 190-370, 160-280, 140-280, 222-382, 220-380, or 230-380 hours.

2. <u>Degradation of Nitrogen-Containing Compounds</u>

The frequencies of EMFs useful for degrading nitrogen-containing compounds range from 10 MHz to 10,000 MHz, e.g., from 5520 MHz to 5540

MHz (e.g., 5521 to 5538 MHz). Exemplary frequencies are 5521, 5522, 5523, 5524, 5525, 5526, 5527, 5528, 5529, 5530, 5531, 5532, 5533, 5534, 5535, 5536, 5537, 5538, 5539, and 5540 MHz. The field strength of the electric field useful for this water treatment function ranges from about 0.5 to 360 mV/cm, for example, from 90-360 mV/cm or 120-340 mV/cm. Exemplary field strengths are 125, 148, 326, and 350 mV/cm.

By way of example, the yeast cells can be cultured in a first series of alternating electric fields each having a frequency in the range of 5521 to 5538 MHz and a field strength in the range of 90 to 360 mV/cm. The yeast cells are exposed to each EMF for about 25 hours. After the first series of culturing, the resultant yeast cells are further incubated in a second series of alternating electric fields for a total of 30 to 128 hours. It may be preferred that the frequencies in the second series of alternating electric fields are identical to those of the first series in sequence and the field strengths in the second series are increased to a higher level within the range of 90 to 360 mV/cm.

25

30

Although the yeast cells can be activated after even a few hours of culturing in the presence of an EMF, it may be preferred that the activated yeast

cells be allowed to multiply and grow in the presence of the EMF(s) for a total of 228-424 or 208-320 hours.

3. <u>Degradation of Environmental Toxins</u>

15

The frequencies of EMFs useful for degrading environmental toxins 5 range from 5 MHz to 1000 MHz, e.g., 70-100, 126-142, 52-70, 30-50, 200-220, 213-229, 133-151, 145-162, 156-176, 163-183, 175-191, 183-205, 114-128, 244-264, 252-278, or 220-250 MHz. Exemplary frequencies are 70, 72, 74, 76, 78, 80, 82, 84, 86, 88, 90, 92, 94, 96, 98, or 100 MHz. The field strength of the electric field useful for this water treatment function ranges from about 0.5 to 350 mV/cm, for example, from about 20-320 mV/cm (e.g., 30-220, 30-230, 30-250, 90-280, 80-280, 100-200, 110-280, 100-220, 116-225, 120-280, 90-190, 100-190, 160-300, 120-300, 200-300, or 130-310 mV/cm). Exemplary field strengths are 48, 89, 93, 98, 103, 107, 110, 112, 124, 126, 130, 133, 138, 168, 200, 202, 213, 219, and 274 mV/cm.

By way of example, the yeast cells can be first cultured in a series of alternating electric fields each having a frequency in the range of 70 to 100 MHz and a field strength in the range of 20 to 320 mV/cm. The yeast cells are exposed to each EMF for about 10 to 25 hours. After culturing in the first series of EMFs, the resultant yeast cells are further incubated in a second series of alternating electric fields for a total of 30 to 120 hours. It may be preferred that the 20 frequencies in the second series of alternating electric fields are identical to those of the first series in sequence and the field strengths in the second series are increased to a higher level within the range of 20 to 320 mV/cm.

Alternatively, the yeast cells can be cultured in a series of alternating electric fields each having a frequency in the range of 70 to 100 MHz and a field strength in the range of 20 to 320 mV/cm. The yeast cells are exposed to each EMF for about 20 to 40 hours. Preferably, the field strength remains the same in the series whereas the frequency progressively increases.

In another embodiment, the yeast cells can be cultured in a first series of alternating electric fields each having a frequency in the range of 126-142, 30 52-70, 30-50, 200-220, 213-229, 133-151, 145-162, 156-176, 163-183, 175-191, 183-205, 114-128, 244-264, 252-278, or 220-250 MHz and a field strength in the

range of 90-280, 80-280, 120-280, 100-200, 110-280, 100-220, 116-225, 90-190, 100-190, 160-300, 120-300, 200-300, or 130-310 mV/cm. The yeast cells are exposed to each EMF for about 10 to 25 hours. After culturing in the first series of EMFs, the resultant yeast cells are further incubated in a second series of alternating electric fields for a total of 30 to 120 hours. It may be preferred that the electric field frequencies in this second series are identical to those in the first series while the field strengths in the second series are increased to a higher level within the above-described ranges. Alternatively, the yeast cells can be cultured in a series of alternating electric fields each having a frequency in the range of 126-10 142, 52-70, 30-50, 200-220, 213-229, 133-151, 145-162, 156-176, 163-183, 175-191, 183-205, 114-128, 244-264, 252-278, or 220-250 MHz and a field strength in the range of 90-280, 80-280, 120-280, 100-200, 110-280, 100-220, 116-225, 90-190, 100-190, 160-300, 120-300, 200-300, or 130-310 mV/cm. The yeast cells are exposed to each EMF for about 20 to 40 hours. Preferably, the field strength remains the same in the series whereas the frequency progressively increases. 15

Although the yeast cells can be activated after even a few hours of culturing in the presence of an EMF, it may be preferred that the activated yeast cells be allowed to multiply and grow in the presence of the EMF(s) for a total of 180-328, 114-244, 80-380, 80-365, 120-350, 90-330, 130-330, 100-280, 110-330, 130-290, 80-290, 110-360, or 110-340 hours.

20

25

4. <u>Conversion of Bio-available Nitrogen in a Culture Medium to</u> Intracellular Nitrogen

The frequencies of EMFs useful for converting bio-available nitrogen in a culture medium to intracellular nitrogen range from about 5 MHz to 5000 MHz, e.g., from about 600 to 700 MHz (e.g., 660 to 680 MHz) or 2160 to 2190 MHz (e.g., 2170 to 2185 MHz). Exemplary frequencies are 662, 664, 666, 668, 670, 672, 674, 676, 678, 2170, 2171, 2172, 2173, 2174, 2175, 2176, 2177, 2178, 2179, 2180, 2181, 2182, 2183, 2184, and 2185 MHz. The field strength of the electric field useful for this water treatment function ranges from about 0.1 to 350 mV/cm, e.g., from 100-350 mV/cm (e.g., 140-320 or 120-290 mV/cm). Exemplary field strengths are 126, 152, 196 and 310 mV/cm.

By way of example, the yeast cells can be cultured in a first series of alternating electric fields each having a frequency in the range of 660 to 680 MHz and a field strength in the range of 100 to 350 mV/cm. The yeast cells are exposed to each EMF for about 18 to 25 hours. After culturing in the first series of EMFs, the resultant yeast cells are further incubated in a second series of alternating electric fields for a total of 25 to 130 hours. It may be preferred that the electric field frequencies of the second series are identical to those of the first series in sequence while the field strengths in the second series are increased to a higher level within the range of 100 to 350 mV/cm.

In another embodiment, the yeast cells can be cultured in a first series of alternating electric fields each having a frequency in the range of 2170 to 2185 MHz and a field strength in the range of 140 to 320 mV/cm. The yeast cells are exposed to each EMF for about 18 to 25 hours. After culturing in the first series of EMF, the resultant yeast cells are further incubated in a second series of alternating electric fields for a total of 25 to 130 hours. It may be preferred that the frequencies in the second series of alternating electric fields are identical to those of the first series in sequence while the field strengths in the second series are increased to a higher level within the range of 140 to 320 mV/cm.

10

25

Although the yeast cells can be activated after even a few hours of culturing in the presence of an EMF, it may be preferred that the activated yeast cells be allowed to multiply and grow in the presence of the EMF(s) for a total of 192-304 or 226-412 hours.

5. <u>Conversion of Bio-available Phosphorus in a Culture Medium to</u> Intracellular Phosphorus

The frequencies of EMFs useful for converting bio-available phosphorus in a culture medium to intracellular phosphorus range from about 5 to 5000 MHz, e.g., from 80 to 440 MHz (e.g., 86-120 MHz or 410-430 MHz). Exemplary frequencies are 86, 88, 90, 92, 94, 96, 98, 100, 102, 104, 106, 108, 110, 112, 114, 116, 118, 120, 410, 411, 412, 413, 414, 415, 416, 417, 418, 419, 420, 421, 422, 423, 424, 425, 426, 427, 428, 429, and 430 MHz. The field strength of the electric field useful for this water treatment function ranges from about 0.5 to

350 mV/cm, e.g., from about 60 to 260 mV/cm. Exemplary field strengths are 68 and 240 mV/cm.

By way of example, the yeast cells can be cultured in a first series of alternating electric fields each having a frequency in the range of 86 to 120 MHz and a field strength in the range of 60 to 260 mV/cm. The yeast cells are exposed to each EMF for about 24 hours. After culturing in the first series of EMFs, the resultant yeast cells are further incubated in a second series of alternating electric fields for a total of 24 to 132 hours. It may be preferred that the frequencies in the second series of alternating electric fields are identical to those of the first series in sequence and the field strengths in the second series are increased to a higher level within the range of 60 to 260 mV/cm.

Although the yeast cells can be activated after even a few hours of culturing in the presence of an EMF, it may be preferred that the activated yeast cells be allowed to multiply and grow in the presence of the EMF(s) for a total of 228-368 hours.

6. Reduction of Odor

10

15

25

30

The frequencies of EMFs useful for reducing odor range from 5 to 5000 MHz, e.g., from 2160 MHz to 2380 MHz (e.g., 2160-2250 MHz or 2280-2380 MHz). Exemplary frequencies are 2160, 2165, 2170, 2175, 2180, 2185, 2190, 2195, 2200, 2205, 2210, 2215, 2220, 2225, 2230, 2235, 2240, 2245, 2250, 2280, 2285, 2290, 2295, 2300, 2305, 2310, 2315, 2320, 2325, 2330, 2335, 2340, 2345, 2350, 2355, 2360, 2365, 2370, 2375, and 2380 MHz.

The field strength of the electric field useful for this water treatment function ranges from about 0.5 to 320 mV/cm, e.g. from 30 to 310 mV/cm (e.g., 40-260, 70-260, 80-250, 90-260, or 140-300 mV/cm). Exemplary field strengths are 98, 240, 250, and 290 mV/cm.

By way of example, the yeast cells can be cultured in a series of alternating electric fields each having a frequency in the range of 2160 to 2250 MHz or 2280 to 2380 MHz and a field strength in the range of 30 to 310 mV/cm. The yeast cells are exposed to each EMF for about 10 to 40 hours. Preferably, the field strength remains the same in the series whereas the frequency progressively increases.

Alternatively, the yeast cells can be cultured in a first series of alternating electric fields each having a frequency in the range of 2280 to 2380 MHz and a field strength in the range of 90 to 260 mV/cm. The yeast cells are exposed to each EMF for about 15 to 20 hours. After culturing in the first series of EMFs, the resultant yeast cells are further incubated in a second series of alternating electric fields for a total of 20 to 50 hours. It may be preferred that the frequencies in the second series of alternating electric fields are identical to those of the first series in sequence and the field strengths in the second series are increased to a higher level within the range of 90 to 260 mV/cm.

Although the yeast cells can be activated after even a few hours of culturing in the presence of an EMF, it may be preferred that the activated yeast cells be allowed to multiply and grow in the presence of the EMF(s) for a total of 70-220, 70-320, 80-310, 85-220, 110-230, or 120-300 hours.

7. Suppression of Growth of Pathogenic Microbes

10

15

20

30

The frequencies of EMFs useful for suppressing the growth of pathogenic microbes range from 30 MHz to 50 MHz. Exemplary frequencies are 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, and 50 MHz. The field strength of the electric field useful for this water treatment function ranges from about 0.5 to 200 mV/cm, e.g., 10 to 180 mV/cm. Exemplary field strengths are 26 and 150 mV/cm.

By way of example, the yeast cells can be cultured in a first series of alternating electric fields each having a frequency in the range of 30 to 50 MHz and a field strength in the range of 10 to 180 mV/cm. The yeast cells are exposed to each EMF for about 12 hours. After culturing in the first series of EMFs, the resultant yeast cells are further incubated under substantially the same conditions in a second series of alternating electric fields for a total of 24 to 96 hours. It may be preferred that the frequencies in the second series of alternating electric fields are identical to those of the first series in sequence and the field strengths in the second series are increased to a higher level within the range of 10 to 180 mV/cm.

Although the yeast cells can be activated after even a few hours of culturing in the presence of an EMF, it may be preferred that the activated yeast

cells be allowed to multiply and grow in the presence of the EMF(s) for a total of 144-272 hours.

8. Suppression of Growth of Algae or Decomposition of Algal Debris
The frequencies of EMFs useful for suppressing the growth of algae

5 range from about 10 to 10,000 MHz, e.g., from about 6340 MHz to 6380 MHz
(e.g., 6352-6370 MHz). Exemplary frequencies are 6352, 6353, 6354, 6355, 6356, 6357, 6358, 6359, 6360, 6361, 6362, 6363, 6364, 6365, 6366, 6367, 6368, 6369, and 6370 MHz. The field strength of the electric field useful for this water treatment function ranges from about 0.5 to 400 mV/cm, e.g., from about 60 to 380 mV/cm (e.g., 70 to 310, 100 to 330, or 120 to 360 mV/cm). Exemplary field strengths are 85, 112, 136, 250, 290, and 337 mV/cm.

In another embodiment, the frequencies of EMFs useful in this invention range from about 4440 to 4470 MHz (e.g., 4452-4470 MHz). Exemplary frequencies are 4452, 4453, 4454, 4455, 4456, 4457, 4458, 4459, 4460, 4461, 4462, 4463, 4464, 4465, 4466, 4467, 4468, 4469, and 4470 MHz. The field strength of the electric field useful in this invention ranges from about 0.5 to 400 mV/cm, e.g., from about 50 to 280 mV/cm. Exemplary field strengths are 127 and 268 mV/cm.

By way of example, the yeast cells can be cultured in a first series of alternating electric fields each having a frequency in the range of 6352 to 6370 MHz and a field strength in the range of 60 to 380 mV/cm. The yeast cells are exposed to each EMF for about 24 hours. After culturing in the first series of EMFs, the resultant yeast cells are further incubated in a second series of alternating electric fields for a total of 56 to 160 hours. It may be preferred that the frequencies in the second series of alternating electric fields are identical to those of the first series in sequence and the field strengths in the second series are increased to a higher level within the range of 60 to 380 mV/cm.

In another embodiment, the yeast cells can be cultured in a first series of alternating electric fields each having a frequency in the range of 4452 to 4470 MHz and a field strength in the range of 50 to 280 mV/cm. The yeast cells are exposed to each EMF for about 32 hours. After culturing in the first series of EMFs, the resultant yeast cells are further incubated in a second series of

30

alternating electric fields for a total of 32 to 192 hours. It may be preferred that the frequencies in the second series of alternating electric fields are identical to those of the first series in sequence and the field strengths in the second series are increased to a higher level within the range of 50 to 280 mV/cm.

Although the yeast cells can be activated after even a few hours of culturing in the presence of an EMF, it may be preferred that the activated yeast cells be allowed to multiply and grow in the presence of the EMF(s) for a total of 256-432 hours.

III. Culture Media

5

Culture media useful in this invention contain sources of nutrients assimilable by yeast cells. In this invention, a culture medium refers to a laboratory culture medium, or liquid or solid waste in need of treatment. Complex carbon-containing substances in a suitable form, such as carbohydrates (e.g., sucrose, glucose, fructose, dextrose, maltose, xylose, cellulose, starches, etc.) and coal, can be the carbon sources for yeast cells. The exact quantity of the carbon sources utilized in the medium can be adjusted in accordance with the other ingredients of the medium. In general, the amount of carbohydrates varies between about 0.1% and 5% by weight of the medium and preferably between about 0.1% and 2%, and most preferably about 1%. These carbon sources can be used individually or in combination. Among the inorganic salts which can be added to the culture medium are the customary salts capable of yielding sodium, potassium, calcium, phosphate, sulfate, carbonate, and like ions. Non-limiting examples of nutrient inorganic salts are (NH₄)₂HPO₄, KH₂PO₄, K₂HPO₄, CaCO₃, MgSO₄, KNO₃, NaCl, and CaSO₄.

IV. Electromagnetic Activation of Yeast Cells

25 <u>Yeast Strains Characterized By A Substantial Increase In Their</u>

<u>Capacity to Degrade Polymeric Compounds</u>

Certain activated yeasts of the invention convert complex, high molecular weight organic polymers into simple molecules such as pentoses and hexoses. When a water treatment composition contains yeast cells of multiple functional groups, the simple molecules produced by the Ca yeasts can then be utilized by yeasts of other functional groups, e.g., as nutrients. Yeasts of this degradation capability are called herein "Ca yeasts." Organic polymers degradable

by these Ca yeasts include, but are not limited to, polysaccharides (e.g., cellulose and hemicellulose), fatty acid, lignin, polyethylene, polypropylene, polyvinyl chloride and polystyrene.

To activate or enhance the innate ability of yeast cells to degrade

5 polymers, these cells can be cultured in an appropriate medium under sterile
conditions at 25°C-30°C (e.g., 28°C) for a sufficient amount of time, e.g., 12-400
hours (e.g., 220-360, 190-370, 160-280, 140-280, 222-382, 220-380, or 230-380
hours), in an alternating electric field or a series of alternating electric fields as
described in Section II(1). An exemplary set-up of the culture process is depicted
in Fig. 1. An exemplary culture medium contains in per 1000 ml of sterile water
the following: 3 g of lignin (no smaller than 120 μm), 3 g of cellulose (no smaller
than 120 mesh), 5 g of crude oil-contaminated water, 0.2 g of NaCl, 0.2 g of
MgSO₄•7H₂O, 0.5 g of CaCO₃•5H₂O, 0.2 g of CaSO₄•2H₂O, and 0.5 g of K₂HPO₄.

Subsequently, the yeast cells can be measured for their ability to

degrade polymers such as polysaccharides using standard methods. In one
embodiment, 1 ml of the prepared yeast culture is inoculated into 30 ml of an
appropriate medium. The culture is incubated at room temperature for 24-72
hours. The amount of simple carbohydrates in the culture can then be determined
by any methods known in the art, including but not limited to chromatography.

Preferably, the amount of simple carbohydrates in the culture is increased by at
least 10 mg for each gram of yeast dry weight.

In another method, wood pulp from paper mills, with a chemical oxygen demand ("COD") level no less than 50,000 mg/L and typically containing lignin, cellulose, and polysaccharides, is used as a substrate. Specifically, the wood pulp is diluted to the following COD concentrations: (1) 100-1,000 mg/L; (2) 1,000-5,000 mg/L; (3) 5,000-10,000 mg/L; and (4) 10,000-50,000 mg/L. The wood pulp solutions are then inoculated with a dry yeast cell preparation at a concentration of 0.2-0.5 g/L, and cultured for 24-120 hours at 10-40°C. The COD levels of the solutions are then measured using standard techniques. The difference between the COD levels before and after 24-120 hours indicates the carbohydrate-degrading activity of the yeast cells. Other methods for determining the polymer-

30

degrading abilities of the activated cells are described below in the working examples.

5

10

15

20

Yeast Strains Characterized By A Substantial Increase In Their
 Capability To Degrade Nitrogen-Containing Compounds

Certain activated yeasts of this invention can degrade complex nitrogen-containing compounds (e.g., proteins, peptides, lipids, and nucleic acids) into simple molecules. Such simple molecules can then be utilized by other yeast cells to support their growth and activities. Yeasts of this degradation capability are called herein "Ni yeasts." Nitrogen-containing compounds degradable by these yeasts include, but are not limited to, proteins, peptides, lipids, and nucleic acids.

To activate or enhance the innate ability of yeast cells to degrade nitrogen-containing compounds, these cells can be cultured in an appropriate medium under sterile conditions at 25°C-30°C (e.g., 28°C) for a sufficient amount of time, e.g., 12-450 hours (e.g., 228-424 or 208-320 hours) in an alternating electric field or a series of alternating electric fields as described in Section II (2). An exemplary set-up of the culture process is depicted in Fig. 1. An exemplary medium contains in per 1000 ml of sterile water: 12 g of soluble starch, 1.2 g of beef protein, 1.2 g of lecithin, 1.2 g of NaCl, 0.2 g of MgSO₄•7H₂O, 3 g of CaCO₃•5H₂O, 0.3 g of CaSO₄•2H₂O, and 0.2 g of K₂HPO₄.

Subsequently, the yeast cells can be measured for their ability to degrade nitrogen-containing compounds using standard methods. In an exemplary method, beef protein and plant protein are used as substrates for the yeast cells. The proteins are added to sterile water to obtain solutions with the following COD concentrations: (1) 100-1,000 mg/L; (2) 1,000-5,000 mg/L; (3) 5,000-10,000 mg/L; and (4) 10,000-50,000 mg/L. The protein solutions are then inoculated with a dry yeast cell preparation at a concentration of 0.3-0.5 g/L, and cultured for 24-72 hours at 10-40°C. The COD levels of the solutions are then measured using standard techniques. The difference between the COD levels before and after 24-72 hours indicates the yeast cells' ability to degrade the nitrogen-containing compounds. Other methods for determining the activated yeast cells' ability to degrade the nitrogen-containing examples.

3. Yeast Strains Characterized By A Substantial Increase In Their

Capability To Degrade Environmental Toxins

Certain activated yeasts of this invention can degrade environmentally harmful toxins, e.g., chemical compounds such as fertilizers, organic solvents, detergents, and antibiotics, into harmless simple molecules. Such yeasts are called herein "Ch yeasts." These compositions are most efficient in degrading compounds having a molecular weight of 180 to 28,000 daltons.

To activate or enhance the innate ability of yeast cells to degrade environmental toxins, the cells can be cultured in an appropriate medium under sterile conditions at 25°C-30°C (e.g., 28°C) for a sufficient amount of time, e.g., 12-400 hours (e.g., 180-328, 114-244, 80-380, 80-365, 120-350, 90-330, 130-330, 100-280, 110-330, 130-290, 80-290, 110-360, or 110-340 hours), in an alternating electric field or a series of alternating electric fields as described in Section II (3).

An exemplary medium contains in per 400 ml of sterile water: 8 g of sludge, 0.2 g of NaCl, 0.2 g of MgSO₄•7H₂O, 0.5 g of CaCO₃•5H₂O, 0.2 g of CaSO₄•2H₂O, 0.5 g of K₂HPO₄, 1.5 g of peptone, and 600 ml of sludge extract. The sludge extract is prepared as follows: 500 g of sludge known to be polluted by harmful chemicals is mixed and incubated with 600 ml of sterile water at 30-37°C for 24 hours. The sludge mix is then filtered to obtain sludge extract.

- 20 Subsequently, the yeast cells can be measured for their ability to degrade the various chemicals using standard methods.
 - 4. Yeast Strains Characterized By A Substantial Increase In Their

 Capability To Convert Bio-available Nitrogen In A Culture Medium

 To Intracellular Nitrogen
- 25 Certain activated yeasts of this invention can convert biologically available or assimilable nitrogen in a culture medium, such as waste water, into their own biomass, i.e., intracellular nitrogen. Such yeasts are called herein "N-C yeasts." Biologically available nitrogen convertible by these yeasts includes, but is not limited to, NH₄⁺, NO₃ and NO₂, other water-soluble inorganic nitrogen-containing compounds, and organic nitrogen-containing compounds. Biologically available nitrogen in waste water causes undesired eutrophication of water bodies in the world.

To activate or enhance the innate ability of yeast cells to convert biologically available nitrogen in a culture medium into intracellular nitrogen, the cells can be cultured in an appropriate medium under sterile conditions at 25°C-30°C (e.g., 28°C) for a sufficient amount of time, e.g., 12-420 hours (e.g., 192-304 or 226-412 hours), in an alternating electric field or a series of alternating electric fields as described in Section II (4). An exemplary medium contains in per 1000 ml of sterile water: 12 g of sucrose, 4 g of NH₄NO₂, 0.25 g of NaCl, 0.25 g of MgSO₄•7H₂O, 3.5 g of CaCO₃•5H₂O, 0.5 g of CaSO₄•2H₂O, and 0.2 g of K₂HPO₄.

Subsequently, the yeast cells can be measured for their ability to convert available nitrogen to intracellular nitrogen using standard methods. In an exemplary method, waste water from a nitrogen fertilizer manufacturer containing high levels of ammonium salts and/or nitrate or nitride salts is mixed with distilled water to achieve the following COD concentrations: (1) 100-1,000 mg/L; (2) 1,000-5,000 mg/L; (3) 5,000-10,000 mg/L; and (4) 10,000-50,000 mg/L. The solutions are then inoculated with dry yeast cell preparation, at a concentration of 0.2-0.6 g/L, and cultured for 24-48 hours at 10-40°C. The COD levels of the solutions are then measured using standard techniques. The difference between the COD levels before and after 24-48 hours indicates the nitrogen-converting ability of the yeast cells. Another method for determining nitrogen compound levels in a culture medium is described below in the working examples.

10

20

5. Yeast Strains Characterized By A Substantial Increase In Their
Capability To Convert Bio-available Phosphorus In A Culture
Medium To Intracellular Phosphorus

25 available or assimilable phosphorus in a culture medium, such as waste water, into their own biomass, i.e., intracellular phosphorus. Such yeasts are called herein "P-C yeasts." Biologically available phosphorus convertible by these yeasts includes, but is not limited to, PO₄³⁻, H₃PO₄, HPO₄², H₂PO₄, other water-soluble inorganic phosphorus-containing compounds, and organic phosphorus-containing compounds. Biologically available phosphorus in waste water causes undesired eutrophication of water bodies in the world.

PCT/GB01/05439 WO 02/070682

To activate or enhance the innate ability of yeast cells to convert biologically available phosphorus into intracellular phosphorus, these cells can be cultured in an appropriate medium under sterile conditions at 25°C-30°C, e.g., 28°C, for a sufficient amount of time, e.g., 12-400 hours (for example, 228-368 hours) in an alternating electric field or a series of alternating electric fields as described in Section II (5). An exemplary culture medium contains in per 1000 ml of sterile water: 10 g of sucrose, 3 g of (NH₄)H₂PO₄ (or other biologically available phosphorus), 1.2 g of NaCl, 0.2 g of MgSO₄•7H₂O, 3 g of CaCO₃•5H₂O, 0.3 g of CaSO₄•2H₂O, 0.3 g of KNO₃, and 0.5 g of yeast extract.

10

20

25

30

Subsequently, the yeast cells can be measured for their ability to convert biologically available phosphorus to intracellular phosphorus using standard methods, such as using ultraviolet spectrophotometry or the chemical oxygen demand ("COD") method. In an exemplary method, waste water from a phosphorus fertilizer manufacturer containing high levels of HPO₄²⁻, H₂PO₄⁻, and/or 15 H₃PO₄ is mixed with distilled water to achieve the following COD concentrations: (1) 100-1,000 mg/L; (2) 1,000-5,000 mg/L; (3) 5,000-10,000 mg/L; and (4) 10,000-50,000 mg/L. The solutions are then inoculated with a dry yeast cell preparation at a concentration of 0.2-0.6 g/L, and cultured for 24-48 hours at 10-40°C. The COD levels of the solutions are then measured using standard techniques. The difference between the COD levels before and after 24-48 hours indicates the phosphorus converting activity of the yeast cells. Another method for determining the phosphorus-converting abilities of the activated cells is described in the working examples, infra.

> Yeast Strains Characterized By A Substantial Increase In Their 6. Capability To Reduce Odor

Certain activated yeasts of this invention reduce odor by lowering the concentration of malodorous materials. These yeasts are called "O" yeasts herein. Malodorous materials include, but are not limited to, hydrogen sulfide, ammonium sulfide, other sulfur-containing compounds, ammonia, indole, methylindoles, p-cresol, amines such as methylamine, dimethylamine and trimethylamine, and odorous organic acids, such as carboxylic acids, e.g., formic acid, acetic acid, propanoic acid and butyric acid, and other volatile fatty acids.

To activate or enhance the innate ability of yeast cells to reduce odor, these cells can be cultured in an appropriate medium under sterile conditions at 25°C-30°C, e.g., 28°C, for a sufficient amount of time, e.g., 12-350 hours (for example, 70-220, 70-320, 80-310, 85-220, 110-230, or 120-300 hours), in an alternating electric field or a series of alternating electric fields as described in Section II (6).

An exemplary culture medium contains in per 1000 ml of sewage water (containing malodorous materials): 0.2 g of NaCl, 0.2 g of MgSO₄•7H₂O, 0.5 g of CaCO₃•5H₂O, 0.2 g of CaSO₄•2H₂O, and 0.5 g of K₂HPO₄.

10

15

Subsequently, the yeast cells can be measured for their ability to reduce odor. Various methods and techniques are known to measure the intensity of an odor, including but not limited to gas chromatography, HPLC, and mass spectrometry. A reduction of the intensity of the odor of malodorous materials can also be determined subjectively. One subjective measurement of odor intensity is to measure the dilution necessary so that the odor is imperceptible or doubtful to a human or animal test panel. Any methods and techniques for objectively or subjectively determining the intensity of an odor can be used to monitor the ability of the yeast compositions to reduce odor.

In an exemplary method, sewage water containing about 2 g/L

20 methylamine/dimethylamine/trimethylamine, 1 g/L indole, 2 g/L p-cresol, 1 g/L

hydrogen sulfide, 2 g/L acetic acid and/or 1 g/L ammonia is used as a substrate.

The sewage is inoculated with a dry yeast cell preparation, at a concentration of

0.2-0.6 g/L, and cultured for 24 hours at 10-35°C. The level of the malodorous

chemical is measured by gas chromatography. The difference between the levels of

the above-mentioned malodorous components before and after 24 hours indicates
the odor-reducing ability of the yeast cells.

7. Yeast Strains Characterized By A Substantial Increase In Their

Capability To Suppress Growth of Pathogenic Microbes

Certain activated yeasts of this invention can suppress the natural

proliferation of pathogenic microbes. Such yeasts are called herein "Pa yeasts."

Normally, in the presence of ample nutrients, the number of pathogenic microbes

would increase naturally over a period of time. These pathogenic microbes

include, but are not limited to, bacteria such as those belonging to the Escherichia, Salmonella, Shigella, Mycobacterium, Staphylococcus, Bacillus, Streptococcus and Diplococcus genera.

To activate or enhance the innate ability of yeast cells to suppress the growth of pathogenic microbes, these yeast cells can be cultured in an appropriate medium under sterile conditions at 25°C-30°C, e.g., 28°C, for a sufficient amount of time, e.g., 12-300 hours (for example, 144-272 hours), in an alternating electric field or a series of alternating electric fields as described in Section II (7).

5

10

20

25

30

An exemplary culture medium is made by mixing 400 ml of sterile water, 8 g of soluble starch, 5 g of sucrose, 0.2 g of NaCl, 0.2 g of MgSO₄•7H₂O, 0.5 g of CaCO₃•5H₂O, 0.2 g of CaSO₄•2H₂O, 0.5 g of K₂HPO₄, 1.5 g of peptone, and 600 ml of sludge extract containing pathogenic microbes.

Subsequently, the yeast cells can be measured for their ability to

suppress the growth of pathogenic microbes using standard methods known in the
art for counting microorganisms, such as optical density, plating out dilutions on
solid media for counting, or counting individual cells under a microscope. Stains
may be applied to distinguish or identify different strains or species of
microorganisms present in a sample, or to determine their viability.

In one exemplary method, sewage containing more than 10° cells/ml Gram-positive Escherichia coli, 10° cells/ml Salmonella, and 10° cells/ml Shigella dysenteriae is used as a substrate. The sewage is inoculated with a dry yeast cell preparation at a concentration of 0.3-0.6 g/L, and cultured for 24 hours at 10-40°C. The difference between the numbers of the above-mentioned live bacteria before and after 24 hours indicates the pathogen-suppressing capacity of the yeast cells.

8. Yeast Strains Characterized By A Substantial Increase In Their

Capability To Suppress Algae Growth Or Decompose Algal Debris

Eutrophication causes overgrowth of harmful algae, which

dramatically decrease the level of dissolved oxygen in water and adversely affect
the aquatic ecosystem. In addition, debris of these algae deposit on sediment,
where oxygen levels are low, and thus cannot be effectively decomposed by natural
microorganisms. Non-decomposed algal debris provide nutrients for further algal

5

15

20

30

growth, generating a vicious cycle of algal pollution. Certain activated yeasts of this invention can prevent or reduce such pollution by inhibiting the proliferation of algae and/or by decomposing algal debris. Such yeasts are called herein "Al yeasts." Algae of this invention include, but are not limited to, green, blue, and red algae.

To activate or enhance the innate ability of yeast cells to suppress algae growth or decompose algal debris, the yeast cells can be cultured in an appropriate medium under sterile conditions at 25°C-30°C, e.g., 28°C, for a sufficient amount of time, e.g., 12-450 hours (for example, 256-432 hours), in an alternating electric field or a series of alternating electric fields as described in Section II (8).

An exemplary culture medium is made by mixing 1000 ml of distilled water with 6 g of dehydrated algal debris, 0.2 g of NaCl, 0.2 g of MgSO₄•7H₂O, 0.5 g of CaCO₃•5H₂O, 0.2 g of CaSO₄•2H₂O, and 0.5 g of K₂HPO₄. The dehydrated algae debris is prepared by centrifuging surface water (e.g., from a pond) containing a large amount of blue and green algae at 1000 g for 20 minutes, and placing the pellet under vacuum for 48 hours.

Subsequently, the yeast cells can be measured for their ability to suppress the growth of algae or decompose algal debris using standard methods known in the art, such as counting individual cells. In one exemplary method, surface water (from e.g., a river, pond, or lake) containing more than 10¹⁰ live blue algae cells/ml and more than 10¹⁰ live green algae cells/ml is inoculated with a dry yeast cell preparation at a concentration of 0.2-0.6 g/L, and cultured for 24-72 hours at 15-42°C. The difference between the numbers of the above-mentioned live algae before and after 24-72 hours indicates the algae-suppressing or decomposing capacity of the yeast cells.

The culturing process of the yeast cells of the present invention may preferably be conducted under conditions in which the concentration of dissolved oxygen is between 0.025 to 0.8 mol/m³, preferably 0.4 mol/m³. The oxygen level can be controlled by, for example, stirring and/or bubbling.

Essentially the same protocol as described above can be used to grow activated yeast cells of the eight water treatment functions. To initiate the

process, each 100 ml of culture medium is inoculated with the activated yeast cells at a density of 10²-10⁶ cells/ml, preferably 10⁵-10⁶ cells/ml, most preferably 3 x 10⁵ cells/ml. The culturing process is carried out at about 20-40°C, preferably about 25-28°C, for 48-96 hours. The process can be scaled up or down according to needs. For an industrial scale of production, seventy-five liters of a sterile culture medium are inoculated with the yeast cells. This culture medium consists of 10 L of the culture medium described above for this particular yeast functional group, 30 kg of starch, and 65 L of distilled water. At the end of the culturing process, the yeast cells may preferably reach a concentration of 2 x10¹⁰ cells/ml. The cells are recovered from the culture by various methods known in the art, and stored at about 15-20°C. The yeast should be dried within 24 hours and stored in powder form.

V. Acclimatization of Yeast Cells To Waste Environment

15

20

25

30

In yet another embodiment of the invention, the activated yeast cells may also be cultured under certain conditions so as to acclimatize the cells to a particular type of waste. This process can be applied to each yeast cell component separately or to a mixture of yeast cell components. This acclimatization process results in better growth and survival of the yeasts in a particular waste environment.

To achieve this, the yeast cells of a given function are mixed with waste from a particular source at 106 to 108 cells (e.g., 107 cells) per 1000 ml. The yeast cells are then exposed to an alternating electric field having a frequency specific for this function as described in Sections II and IV, *supra*. The strength of the electric field can be about 100 to 400 mV/cm (e.g., 120-250 mV/cm). The culture is incubated at temperatures that cycle between about 5°C to about 45°C at a 5°C increment. For example, in a typical cycle, the temperature of the culture may start at 5°C and be kept at this temperature for about 1-2 hours, then adjusted up to 10°C and kept at this temperature for 1-2 hours, then adjusted to 15°C and kept at this temperature for about 1-2 hours, and so on and so forth, until the temperature reaches 45°C. Then the temperature is brought down to 40°C and kept at this temperature for about 1-2 hours, and then to 35°C and kept at this temperature for about 1-2 hours, and so on and so forth, until the temperature for about 1-2 hours, and so on and so forth, until the temperature

returns to 5°C. The cycles are repeated for about 48-96 hours. The resulting yeast cells are then dried and stored at 0-4°C.

VI. Manufacture of the Water Treatment Compositions

The yeast cells of this invention, with the same or different water

treatment functions, can be mixed with an appropriate filler, such as rock powder
and coal ash at the following ratio: 600 L of mixed yeast cell culture at 2 x10¹⁰
cells/ml and 760 kg of filler materials. The mixture is quickly dried at a
temperature below 65°C for 10 minutes in a dryer, and then further dried at a
temperature below 70°C for no more than 30 minutes, so that the water content is
less than 7%. The dried composition is then cooled to room temperature for
packaging:

These dried yeast compositions may be used to treat polluted surface water, sewage, or any other type of liquid or solid waste. By way of example, to treat polluted surface water, a yeast solution may be prepared by adding 1 kg of the dried yeast composition to 30 L of clean water. The yeast solution is then sprayed onto the polluted surface water at about 1-3 L of the solution per square meter of the polluted surface water. To treat sewage or any other type of waste water, a yeast solution may be prepared by adding about 1 kg of the dried yeast composition to 10-30 L of clean water. The yeast solution is incubated at 10-35 °C for 24-48 hours. The resultant yeast solution is then added to the waste water at about 3-20 L of the solution per liter of waste water.

VII. Examples

20

The following examples are meant to illustrate the methods and materials of the present invention. Suitable modifications and adaptations of the described conditions and parameters which are obvious to those skilled in the art are within the spirit and scope of the present invention.

Example 1: Degradation of Lignin

Saccharomyces cerevisiae Hansen AS2.502 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 4243 MHz and a field strength of 95 mV/cm for 10 hours; (2) then to an alternating electric field having a frequency of 4246 MHz and a field strength of 95 mV/cm for

PCT/GB01/05439 WO 02/070682

10 hours; (3) then to an alternating electric field having a frequency of 4253 MHz and a field strength of 95 mV/cm for 30 hours; (4) then to an alternating electric field having a frequency of 4256 MHz and a field strength of 95 mV/cm for 30 hours; (5) then to an alternating electric field having a frequency of 4243 MHz and a field strength of 300 mV/cm for 20 hours; (6) then to an alternating electric field having a frequency of 4246 MHz and a field strength of 300 mV/cm for 20 hours; (7) then to an alternating electric field having a frequency of 4253 MHz and a field strength of 300 mV/cm for 50 hours; and (8) finally to an alternating electric field having a frequency of 4256 MHz and a field strength of 300 mV/cm for 50 hours.

To test the lignin-degrading activity of the cultured cells, industrial waste water containing large amounts of lignin was supplemented with additional lignin to reconstitute a solution containing lignin at 200 mg/L. 0.1 ml of the EMFtreated AS2.502 cells at a concentration higher than 108 cells/ml was added to 100 L of the lignin solution and cultured at 28°C for 48 hours (solution A). One 15 hundred liters of the lignin solution containing the same number of non-treated AS2.502 cells (solution B) or containing no cells (solution C) were used as controls. The COD levels of the solutions were measured. Alternatively, the solutions were examined using HPLC. The results showed that after 48 hours of incubation, the lignin concentration in solution A decreased more than 12% relative to solution C. In contrast, the lignin concentration of solution B showed no significant change relative to solution C.

Example 2: Degradation of Cellulose

10

Saccharomyces cerevisiae Hansen AS2.516 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 4243 MHz and a field strength of 110 mV/cm for 25 hours; (2) then to an alternating electric field having a frequency of 4248 MHz and a field strength of 110 mV/cm for 25 hours; (3) then to an alternating electric field having a frequency of 4253 MHz and a field strength of 110 mV/cm for 25 hours; (4) then to an alternating electric field having a frequency of 4258 MHz and a field strength of 110 mV/cm for 25 hours; (5) then to an alternating electric field having a frequency of 4243 MHz and a field strength of 280 mV/cm for 20 hours; (6) then to an

alternating electric field having a frequency of 4248 MHz and a field strength of 280 mV/cm for 30 hours; (7) then to an alternating electric field having a frequency of 4253 MHz and a field strength of 280 mV/cm for 30 hours; and (8) finally to an alternating electric field having a frequency of 4258 MHz and a field strength of 280 mV/cm for 10 hours.

To test the cellulose-degrading activity of the cultured cells, waste water containing cellulose was supplemented with additional cellulose to reconstitute a solution containing cellulose at 200 mg/L. 0.1 ml of the EMF-treated AS2.516 cells at a concentration higher than 108 cells/ml was added to 100 L of the cellulose solution and cultured at 28°C for 48 hours (solution A). One hundred liters of the cellulose solution containing the same number of non-treated AS2.516 cells (solution B) or containing no cells (solution C) were used as controls. The COD levels of the solutions were measured. Alternatively, the solutions were examined using HPLC. The results showed that after 48 hours of incubation, the cellulose concentration in solution A decreased more than 22% relative to solution C. In contrast, the cellulose concentration of solution B showed no significant change relative to solution C.

Example 3: Degradation of Hemicellulose

the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 4245 MHz and a field strength of 87 mV/cm for 15 hours; (2) then to an alternating electric field having a frequency of 4250 MHz and a field strength of 87 mV/cm for 15 hours; (3) then to an alternating electric field having a frequency of 4255 MHz and a field strength of 87 mV/cm for 15 hours; (4) then to an alternating electric field having a frequency of 4260 MHz and a field strength of 87 mV/cm for 15 hours; (5) then to an alternating electric field having a frequency of 4245 MHz and a field strength of 250 mV/cm for 25 hours; (6) then to an alternating electric field having a frequency of 4250 MHz and a field strength of 250 mV/cm for 25 hours; (7) then to an alternating electric field having a frequency of 4250 MHz and a field strength of 250 mV/cm for 25 hours; and (8) finally to an alternating electric field having a frequency of 4260 MHz and a field strength of 250 mV/cm for 25 hours.

To test the hemicellulose-degrading activity of the cultured cells, waste water was supplemented with hemicellulose to reconstitute a solution containing hemicellulose at 200 mg/L. 0.1 ml of the EMF-treated AS2.409 cells at a concentration higher than 10⁸ cells/ml was added to 100 L of the hemicellulose solution and cultured at 28°C for 48 hours (solution A). One hundred liters of the hemicellulose solution containing the same number of non-treated AS2.409 cells (solution B) or containing no cells (solution C) were used as controls. The COD levels of the solutions were measured. Alternatively, the solutions were examined using HPLC. The results showed that after 48 hours of incubation, the hemicellulose concentration in solution A decreased more than 24% relative to solution C. In contrast, the hemicellulose concentration of solution B showed no significant change relative to solution C.

Example 4: Degradation of Polyethylene

15

20

25

Saccharomyces cerevisiae Hansen AS2.430 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 4244 MHz and a field strength of 78 mV/cm for 15 hours; (2) then to an alternating electric field having a frequency of 4247 MHz and a field strength of 78 mV/cm for 15 hours; (3) then to an alternating electric field having a frequency of 4254 MHz and a field strength of 78 mV/cm for 15 hours; (4) then to an alternating electric field having a frequency of 4258 MHz and a field strength of 78 mV/cm for 15 hours; (5) then to an alternating electric field having a frequency of 4244 MHz and a field strength of 250 mV/cm for 20 hours; (6) then to an alternating electric field having a frequency of 4247 MHz and a field strength of 250 mV/cm for 20 hours; (7) then to an alternating electric field having a frequency of 4254 MHz and a field strength of 250 mV/cm for 20 hours; and (8) finally to an alternating electric field having a frequency of 4258 MHz and a field strength of 250 mV/cm for 20 hours.

To test the polyethylene-degrading activity of the cultured cells, industrial waste water containing polyethylene was supplemented with additional polyethylene (≥80 mesh) to reconstitute a solution containing polyethylene at 200 mg/L. 0.1 ml of the EMF-treated AS2.430 cells at a concentration higher than 10⁸ cells/ml was added to 100 L of the polyethylene solution and cultured at 28°C for

48 hours (solution A). One hundred liters of the polyethylene solution containing the same number of non-treated AS2.430 cells (solution B) or containing no cells (solution C) were used as controls. The COD levels of the solutions were measured. Alternatively, the solutions were examined using HPLC. The results showed that after 48 hours of incubation, the polyethylene concentration in solution A decreased more than 15% relative to solution C. In contrast, the polyethylene concentration of solution B showed no significant change relative to solution C.

Example 5: Degradation of Polypropylene

10

15

20

Saccharomyces cerevisiae Hansen AS2.453 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 4242 MHz and a field strength of 108 mV/cm for 25 hours; (2) then to an alternating electric field having a frequency of 4248 MHz and a field strength of 108 mV/cm for 25 hours; (3) then to an alternating electric field having a frequency of 4253 MHz and a field strength of 108 mV/cm for 25 hours; (4) then to an alternating electric field having a frequency of 4259 MHz and a field strength of 108 mV/cm for 25 hours; (5) then to an alternating electric field having a frequency of 4242 MHz and a field strength of 300 mV/cm for 36 hours; (6) then to an alternating electric field having a frequency of 4248 MHz and a field strength of 300 mV/cm for 36 hours; (7) then to an alternating electric field having a frequency of 4253 MHz and a field strength of 300 mV/cm for 25 hours; and (8) finally to an alternating electric field having a frequency of 4259 MHz and a field strength of 300 mV/cm for 25 hours.

To test the polypropylene-degrading activity of the cultured cells, industrial waste water containing polypropylene was supplemented with additional polypropylene (≥80 mesh) to reconstitute a solution containing polypropylene at 200 mg/L. 0.1 ml of the EMF-treated AS2.453 cells at a concentration higher than 10⁸ cells/ml was added to 100 L of the polypropylene solution and cultured at 28°C for 48 hours (solution A). One hundred liters of the polypropylene solution containing the same number of non-treated AS2.453 cells (solution B) or containing no cells (solution C) were used as controls. The COD levels of the solutions were measured. Alternatively, the solutions were examined using HPLC.

The results showed that after 48 hours of incubation, the polypropylene concentration in solution A decreased more than 19% relative to solution C. In contrast, the polypropylene concentration of solution B showed no significant change relative to solution C.

Example 6: Degradation of Polyvinyl chloride

5

10

15

20

25

Saccharomyces cerevisiae Hansen AS2.463 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 4246 MHz and a field strength of 90 mV/cm for 30 hours; (2) then to an alternating electric field having a frequency of 4250 MHz and a field strength of 90 mV/cm for 30 hours; (3) then to an alternating electric field having a frequency of 4256 MHz and a field strength of 90 mV/cm for 30 hours; (4) then to an alternating electric field having a frequency of 4260 MHz and a field strength of 90 mV/cm for 30 hours; (5) then to an alternating electric field having a frequency of 4246 MHz and a field strength of 240 mV/cm for 25 hours; (6) then to an alternating electric field having a frequency of 4256 MHz and a field strength of 240 mV/cm for 25 hours; (7) then to an alternating electric field having a frequency of 4260 MHz and a field strength of 240 mV/cm for 25 hours; and (8) finally to an alternating electric field having a frequency of 4260 MHz and a field strength of 240 mV/cm for 25 hours.

To test the polyvinyl chloride-degrading activity of the cultured cells, industrial waste water containing polyvinyl chloride was supplemented with additional polyvinyl chloride (≥80 mesh) to reconstitute a solution containing polyvinyl chloride at 200 mg/L. 0.1 ml of the EMF-treated AS2.463 cells at a concentration higher than 10⁸ cells/ml was added to 100 L of the polyvinyl chloride solution and cultured at 28°C for 48 hours (solution A). One hundred liters of the polyvinyl chloride solution containing the same number of non-treated AS2.463 cells (solution B) or containing no cells (solution C) were used as controls. The COD levels of the solutions were measured. Alternatively, the solutions were examined using HPLC. The results showed that after 48 hours of incubation, the polyvinyl chloride concentration in solution A decreased more than 27% relative to solution C. In contrast, the lignin concentration of solution B showed no significant change relative to solution C.

Example 7: Degradation of Polystyrene

15

Saccharomyces cerevisiae Hansen AS2.467 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 5 4243 MHz and a field strength of 82 mV/cm for 25 hours; (2) then to an alternating electric field having a frequency of 4246 MHz and a field strength of 82 mV/cm for 25 hours; (3) then to an alternating electric field having a frequency of 4251 MHz and a field strength of 82 mV/cm for 25 hours; (4) then to an alternating electric field having a frequency of 4257 MHz and a field strength of 82 mV/cm for 25 hours; (5) then to an alternating electric field having a frequency of 4243 MHz and a field strength of 245 mV/cm for 45 hours; (6) then to an alternating electric field having a frequency of 4246 MHz and a field strength of 245 mV/cm for 45 hours; (7) then to an alternating electric field having a frequency of 4251 MHz and a field strength of 245 mV/cm for 20 hours; and (8) finally to an alternating electric field having a frequency of 4257 MHz and a field strength of 245 mV/cm for 20 hours.

To test the polystyrene-degrading activity of the cultured cells, industrial waste water containing polystyrene was supplemented with additional polystyrene (≥80 mesh) to reconstitute a solution containing polystyrene at 200 mg/L. 0.1 ml of the EMF-treated AS2.467 cells at a concentration higher than 10⁸ 20 cells/ml was added to 100 L of the polystyrene solution and cultured at 28°C for 48 hours (solution A). One hundred liters of the polystyrene solution containing the same number of non-treated AS2.467 cells (solution B) or containing no cells (solution C) were used as controls. The COD levels of the solutions were measured. Alternatively, the solutions were examined using HPLC. The results showed that after 48 hours of incubation, the polystyrene concentration in solution A decreased more than 21% relative to solution C. In contrast, the polystyrene concentration of solution B showed no significant change relative to solution C.

Example 8: Degradation of Animal Protein

Saccharomyces cerevisiae Hansen AS2.452 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 5521 MHz and a field strength of 148 mV/cm for 25 hours; (2) then to an

alternating electric field having a frequency of 5526 MHz and a field strength of 148 mV/cm for 25 hours; (3) then to an alternating electric field having a frequency of 5533 MHz and a field strength of 148 mV/cm for 25 hours; (4) then to an alternating electric field having a frequency of 5536 MHz and a field strength of 148 mV/cm for 25 hours; (5) then to an alternating electric field having a frequency of 5521 MHz and a field strength of 350 mV/cm for 32 hours; (6) then to an alternating electric field having a frequency of 5526 MHz and a field strength of 350 mV/cm for 32 hours; (7) then to an alternating electric field having a frequency of 5533 MHz and a field strength of 350 mV/cm for 32 hours; and (8) finally to an alternating electric field having a frequency of 5536 MHz and a field strength of 350 mV/cm for 32 hours.

To test the ability of the EMF-treated AS2.452 cells to degrade animal protein, waste water containing animal protein was supplemented with beef extracts to reconstitute a solution containing protein at 200 mg/L. 0.1 ml of the EMF-treated AS2.452 cells at a concentration higher than 10⁸ cells/ml was added to 150 L of the protein solution and cultured at 28°C for 72 hours (solution A). One hundred and fifty liters of the protein solution containing the same number of non-treated AS2.452 cells (solution B) or containing no cells (solution C) were used as controls. After 72 hours of incubation, the protein solutions were examined using HPLC. The results showed that after 72 hours of incubation, the protein concentration of solution A decreased more than 32% relative to solution C. In contrast, the protein concentration of solution B showed no significant change relative to solution C.

Example 9: Degradation of Plant Protein

20

25

30

Saccharomyces cerevisiae Hansen AS2.423 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 5524 MHz and a field strength of 125 mV/cm for 24 hours; (2) then to an alternating electric field having a frequency of 5528 MHz and a field strength of 125 mV/cm for 24 hours; (3) then to an alternating electric field having a frequency of 5532 MHz and a field strength of 125 mV/cm for 24 hours; (4) then to an alternating electric field having a frequency of 5538 MHz and a field strength of

125 mV/cm for 24 hours; (5) then to an alternating electric field having a frequency of 5524 MHz and a field strength of 326 mV/cm for 28 hours; (6) then to an alternating electric field having a frequency of 5528 MHz and a field strength of 326 mV/cm for 28 hours; (7) then to an alternating electric field having a frequency of 5532 MHz and a field strength of 326 mV/cm for 28 hours; and (8) finally to an alternating electric field having a frequency of 5538 MHz and a field strength of 326 mV/cm for 28 hours.

To test the ability of the EMF-treated AS2.423 cells to degrade plant protein, waste water containing plant protein was supplemented with soy protein to reconstitute a solution containing protein at 200 mg/L. 0.1 ml of the EMF-treated AS2.423 cells at a concentration higher than 10⁸ cells/ml was added to 150 L of the protein solution and cultured at 28°C for 72 hours (solution A). One hundred and fifty liters of the protein solution containing the same number of non-treated AS2.423 cells (solution B) or containing no cells (solution C) were used as controls. After 72 hours of incubation, the protein solutions were examined using HPLC. The results showed that after 72 hours of incubation, the protein concentration of solution A decreased more than 36% relative to solution C. In contrast, the protein concentration of solution B showed no significant change relative to solution C.

Example 10: Degradation of Penicillin

15

20

Saccharomyces willianus Saccardo AS2.293 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 77 MHz and a field strength of 48 mV/cm for 15 hours; (2) then to an alternating electric field having a frequency of 83 MHz and a field strength of 48 mV/cm for 15 hours; (3) then to an alternating electric field having a frequency of 90 MHz and a field strength of 48 mV/cm for 15 hours; (4) then to an alternating electric field having a frequency of 96 MHz and a field strength of 48 mV/cm for 15 hours; (5) then to an alternating electric field having a frequency of 77 MHz and a field strength of 200 mV/cm for 30 hours; (6) then to an alternating electric field having a frequency of 83 MHz and a field strength of 200 mV/cm for 30 hours; (7) then to an alternating electric field having a frequency of 90 MHz and a field strength of

200 mV/cm for 30 hours; and (8) finally to an alternating electric field having a frequency of 96 MHz and a field strength of 200 mV/cm for 30 hours.

To test the ability of the EMF-treated AS2.293 cells to degrade penicillin, waste water from a hospital was supplemented with procainpenzyl penicillin (or cloxacillin) to reconstitute a solution containing penicillin at 100 mg/L. 0.1 ml of the EMF-treated AS2.293 cells at a concentration higher than 108 cells/ml was added to 100 L of the penicillin solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the penicillin solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 24 hours of incubation, the penicillin solutions were examined using HPLC. The results showed that after 24 hours of incubation, the penicillin concentration of solution A decreased more than 23% relative to solution C. In contrast, the penicillin concentration of solution B showed no significant change relative to solution C.

Example 11: Degradation of Aureomycin

15

20

25

30

Saccharomyces cerevisiae Hansen IFFI1063 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 70 MHz and a field strength of 48 mV/cm for 15 hours; (2) then to an alternating electric field having a frequency of 73 MHz and a field strength of 48 mV/cm for 15 hours; (3) then to an alternating electric field having a frequency of 88 MHz and a field strength of 48 mV/cm for 15 hours; (4) then to an alternating electric field having a frequency of 98 MHz and a field strength of 48 mV/cm for 15 hours; (5) then to an alternating electric field having a frequency of 70 MHz and a field strength of 200 mV/cm for 30 hours; (6) then to an alternating electric field having a frequency of 73 MHz and a field strength of 200 mV/cm for 30 hours; (7) then to an alternating electric field having a frequency of 88 MHz and a field strength of 200 mV/cm for 30 hours; and (8) finally to an alternating electric field having a frequency of 98 MHz and a field strength of 200 mV/cm for 30 hours.

To test the ability of the EMF-treated IFFI1063 cells to degrade aureomycin, waste water from a hospital was supplemented with chlortetracycline hydrochloride (one type of aureomycin) to reconstitute a solution containing the

antibiotics at 100 mg/L. 0.1 ml of the EMF-treated IFFI1063 cells at a concentration higher than 108 cells/ml was added to 100 L of the antibiotics solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the antibiotics solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 24 hours of incubation, the antibiotics solutions were examined using HPLC. The results showed that after 24 hours of incubation, the chlortetracycline hydrochloride concentration of solution A decreased more than 31% relative to solution C. In contrast, the chlortetracycline hydrochloride concentration of solution B showed no significant change relative to solution C.

Example 12: Degradation of Oxytetracycline

10

Saccharomyces cerevisiae Hansen IFFI1211 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 70 MHz and a field strength of 48 mV/cm for 15 hours; (2) then to an alternating electric field having a frequency of 74 MHz and a field strength of 48 mV/cm for 15 hours; (3) then to an alternating electric field having a frequency of 88 MHz and a field strength of 44 mV/cm for 15 hours; (4) then to an alternating electric field having a frequency of 98 MHz and a field strength of 48 mV/cm for 15 hours; (5) then to an alternating electric field having a frequency of 70 MHz and a field strength of 200 mV/cm for 30 hours; (6) then to an alternating electric field having a frequency of 74 MHz and a field strength of 200 mV/cm for 30 hours; (7) then to an alternating electric field having a frequency of 88 MHz and a field strength of 200 mV/cm for 30 hours; and (8) finally to an alternating electric field having a frequency of 98 MHz and a field strength of 200 mV/cm for 30 hours.

To test the ability of the EMF-treated IFFI1211 cells to degrade oxytetracycline ("OTC"), waste water from a hospital was supplemented with oxytetracycline hydrochloride to reconstitute a solution containing the antibiotics at 100 mg/L. 0.1 ml of the EMF-treated IFFI1211 cells at a concentration higher than 108 cells/ml was added to 100 L of the antibiotics solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the antibiotics solution containing the same number of non-treated cells (solution B) or containing no cells (solution C)

were used as controls. After 24 hours of incubation, the antibiotics solutions were examined using HPLC. The results showed that after 24 hours of incubation, the OTC concentration of solution A decreased more than 28% relative to solution C. In contrast, the OTC concentration of solution B showed no significant change relative to solution C.

Example 13: Degradation of Doxytetracycline

5

10

15

20

Saccharomyces cerevisiae Hansen IFFI1340 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 71 MHz and a field strength of 48 mV/cm for 15 hours; (2) then to an alternating electric field having a frequency of 73 MHz and a field strength of 48 mV/cm for 15 hours; (3) then to an alternating electric field having a frequency of 77 MHz and a field strength of 48 mV/cm for 15 hours; (4) then to an alternating electric field having a frequency of 88 MHz and a field strength of 48 mV/cm for 15 hours; (5) then to an alternating electric field having a frequency of 71 MHz and a field strength of 200 mV/cm for 30 hours; (6) then to an alternating electric field having a frequency of 73 MHz and a field strength of 200 mV/cm for 30 hours; (7) then to an alternating electric field having a frequency of 77 MHz and a field strength of 200 mV/cm for 30 hours; and (8) finally to an alternating electric field having a frequency of 88 MHz and a field strength of 200 mV/cm for 30 hours; and (8) finally to an alternating electric field having a frequency of 88 MHz and a field strength of 200 mV/cm for 30 hours.

To test the ability of the EMF-treated IFFI1340 cells to degrade doxycycline ("DOTC"), waste water from a hospital was supplemented with DOTC to reconstitute a solution containing the antibiotics at 100 mg/L. 0.1 ml of the EMF-treated IFFI1340 cells at a concentration higher than 108 cells/ml was added to 100 L of the antibiotics solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the antibiotics solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 24 hours of incubation, the antibiotics solutions were examined using HPLC. The results showed that after 24 hours of incubation, the DOTC concentration of solution A decreased more than 33% relative to solution C. In contrast, the DOTC concentration of solution B showed no significant change relative to solution C.

Example 14: Degradation of Tetracycline

Saccharomyces cerevisiae Hansen IFFI1215 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 70 MHz and a field strength of 48 mV/cm for 15 hours; (2) then to an alternating electric field having a frequency of 75 MHz and a field strength of 48 mV/cm for 15 hours; (3) then to an alternating electric field having a frequency of 82 MHz and a field strength of 48 mV/cm for 15 hours; (4) then to an alternating electric field having a frequency of 85 MHz and a field strength of 48 mV/cm for 15 hours; (5) then to an alternating electric field having a frequency of 70 MHz and a field strength of 200 mV/cm for 30 hours; (6) then to an alternating electric field having a frequency of 82 MHz and a field strength of 200 mV/cm for 30 hours; (7) then to an alternating electric field having a frequency of 82 MHz and a field strength of 200 mV/cm for 30 hours; and (8) finally to an alternating electric field having a frequency of 85 MHz and a field strength of 200 mV/cm for 30 hours.

To test the ability of the EMF-treated IFFI1215 cells to degrade tetracycline ("TC"), waste water from a hospital was supplemented with TC to reconstitute a solution containing the antibiotics at 100 mg/L. 0.1 ml of the EMF-treated IFFI1215 cells at a concentration higher than 108 cells/ml was added to 100 L of the antibiotics solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the antibiotics solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 24 hours of incubation, the antibiotics solutions were examined using HPLC. The results showed that after 24 hours of incubation, the TC concentration of solution A decreased more than 26% relative to solution C. In contrast, the TC concentration of solution B showed no significant change relative to solution C.

Example 15: Degradation of Streptomycin

15

Saccharomyces cerevisiae Hansen IFFI1213 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 70 MHz and a field strength of 48 mV/cm for 15 hours; (2) then to an alternating electric field having a frequency of 73 MHz and a field strength of 48 mV/cm for

a field strength of 48 mV/cm for 15 hours; (4) then to an alternating electric field having a frequency of 96 MHz and a field strength of 48 mV/cm for 15 hours; (5) then to an alternating electric field having a frequency of 70 MHz and a field strength of 200 mV/cm for 30 hours; (6) then to an alternating electric field having a frequency of 73 MHz and a field strength of 200 mV/cm for 30 hours; (6) then to an alternating electric field having a frequency of 73 MHz and a field strength of 200 mV/cm for 30 hours; (7) then to an alternating electric field having a frequency of 80 MHz and a field strength of 200 mV/cm for 30 hours; and (8) finally to an alternating electric field having a frequency of 96 MHz and a field strength of 200 mV/cm for 30 hours.

streptomycin, waste water from a hospital was supplemented with streptomycin to reconstitute a solution containing the antibiotics at 100 mg/L. 0.1 ml of the EMF-treated IFFI1213 cells at a concentration higher than 10⁸ cells/ml was added to 100 L of the antibiotics solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the antibiotics solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 24 hours of incubation, the antibiotics solutions were examined using HPLC. The results showed that after 24 hours of incubation, the streptomycin concentration of solution A decreased more than 31% relative to solution C. In contrast, the streptomycin concentration of solution B showed no significant change relative to solution C.

Example 16: Degradation of Kanamycin

10

20

25

Saccharomyces cerevisiae Hansen IFFI1206 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 71 MHz and a field strength of 48 mV/cm for 15 hours; (2) then to an alternating electric field having a frequency of 78 MHz and a field strength of 48 mV/cm for 15 hours; (3) then to an alternating electric field having a frequency of 86 MHz and a field strength of 48 mV/cm for 15 hours; (4) then to an alternating electric field having a frequency of 98 MHz and a field strength of 48 mV/cm for 15 hours; (5) then to an alternating electric field having a frequency of 71 MHz and a field strength of 200 mV/cm for 30 hours; (6) then to an alternating electric field having

a frequency of 78 MHz and a field strength of 200 mV/cm for 30 hours; (7) then to an alternating electric field having a frequency of 86 MHz and a field strength of 200 mV/cm for 30 hours; and (8) finally to an alternating electric field having a frequency of 98 MHz and a field strength of 200 mV/cm for 30 hours.

To test the ability of the EMF-treated IFFI1206 cells to degrade kanamycin, waste water from a hospital was supplemented with kanamycin to reconstitute a solution containing the antibiotics at 100 mg/L. 0.1 ml of the EMF-treated IFFI1206 cells at a concentration higher than 10⁸ cells/ml was added to 100 L of the antibiotics solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the antibiotics solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 24 hours of incubation, the antibiotics solutions were examined using HPLC. The results showed that after 24 hours of incubation, the kanamycin concentration of solution A decreased more than 25% relative to solution C. In contrast, the kanamycin concentration of solution B showed no significant change relative to solution C.

Example 17: Degradation of Erythromycin

5

20

25

30

Saccharomyces cerevisiae Hansen IFFI1211 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 73 MHz and a field strength of 48 mV/cm for 15 hours; (2) then to an alternating electric field having a frequency of 79 MHz and a field strength of 48 mV/cm for 15 hours; (3) then to an alternating electric field having a frequency of 88 MHz and a field strength of 48 mV/cm for 15 hours; (4) then to an alternating electric field having a frequency of 98 MHz and a field strength of 48 mV/cm for 15 hours; (5) then to an alternating electric field having a frequency of 73 MHz and a field strength of 200 mV/cm for 30 hours; (6) then to an alternating electric field having a frequency of 79 MHz and a field strength of 200 mV/cm for 30 hours; (7) then to an alternating electric field having a frequency of 88 MHz and a field strength of 200 mV/cm for 30 hours; and (8) finally to an alternating electric field having a frequency of 98 MHz and a field strength of 200 mV/cm for 30 hours.

To test the ability of the EMF-treated IFFI1211 cells to degrade erythromycin, waste water from a hospital was supplemented with erythromycin to reconstitute a solution containing the antibiotics at 100 mg/L. 0.1 ml of the EMFtreated IFFI1211 cells at a concentration higher than 108 cells/ml was added to 100 5 L of the antibiotics solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the antibiotics solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 24 hours of incubation, the antibiotics solutions were examined using HPLC. The results showed that after 24 hours of incubation, the erythromycin concentration of solution A decreased more than 27% relative to solution C. In contrast, the erythromycin concentration of solution B showed no significant change relative to solution C.

Example 18: Degradation of Spiramycin

10

15

20

25

Saccharomyces cerevisiae Hansen IFFI1210 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 70 MHz and a field strength of 48 mV/cm for 15 hours; (2) then to an alternating electric field having a frequency of 77 MHz and a field strength of 48 mV/cm for 15 hours; (3) then to an alternating electric field having a frequency of 84 MHz and a field strength of 48 mV/cm for 15 hours; (4) then to an alternating electric field having a frequency of 93 MHz and a field strength of 48 mV/cm for 15 hours; (5) then to an alternating electric field having a frequency of 70 MHz and a field strength of 200 mV/cm for 30 hours; (6) then to an alternating electric field having a frequency of 77 MHz and a field strength of 200 mV/cm for 30 hours; (7) then to an alternating electric field having a frequency of 84 MHz and a field strength of 200 mV/cm for 30 hours; and (8) finally to an alternating electric field having a frequency of 93 MHz and a field strength of 200 mV/cm for 30 hours.

To test the ability of the EMF-treated IFFI1210 cells to degrade spiramycin, waste water from a hospital was supplemented with spiramycin to 30 reconstitute a solution containing the antibiotics at 100 mg/L. 0.1 ml of the EMFtreated IFFI1210 cells at a concentration higher than 108 cells/ml was added to 100 L of the antibiotics solution and cultured at 28°C for 24 hours (solution A). One

hundred liters of the antibiotics solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 24 hours of incubation, the antibiotics solutions were examined using HPLC. The results showed that after 24 hours of incubation, the spiramycin concentration of solution A decreased more than 22% relative to solution C. In contrast, the spiramycin concentration of solution B showed no significant change relative to solution C.

Example 19: Degradation of Bacitracin

10

20

25

Saccharomyces cerevisiae Hansen IFFI1290 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 75 MHz and a field strength of 48 mV/cm for 15 hours; (2) then to an alternating electric field having a frequency of 78 MHz and a field strength of 48 mV/cm for 15 hours; (3) then to an alternating electric field having a frequency of 81 MHz and a field strength of 48 mV/cm for 15 hours; (4) then to an alternating electric field having a frequency of 95 MHz and a field strength of 48 mV/cm for 15 hours; (5) then to an alternating electric field having a frequency of 75 MHz and a field strength of 200 mV/cm for 30 hours; (6) then to an alternating electric field having a frequency of 78 MHz and a field strength of 200 mV/cm for 30 hours; (7) then to an alternating electric field having a frequency of 81 MHz and a field strength of 200 mV/cm for 30 hours; and (8) finally to an alternating electric field having a frequency of 95 MHz and a field strength of 200 mV/cm for 30 hours.

To test the ability of the EMF-treated IFFI1290 cells to degrade bacitracin, waste water from a hospital was supplemented with bacitracin to reconstitute a solution containing the antibiotics at 100 mg/L. 0.1 ml of the EMFtreated IFFI1290 cells at a concentration higher than 108 cells/ml was added to 100 L of the antibiotics solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the antibiotics solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 30 24 hours of incubation, the antibiotics solutions were examined using HPLC. The results showed that after 24 hours of incubation, the bacitracin concentration of solution A decreased more than 17% relative to solution C. In contrast, the

bacitracin concentration of solution B showed no significant change relative to solution C.

Example 20: Degradation of Trichloromethane

Saccharomyces cerevisiae Hansen IFFI1413 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 82 MHz and a field strength of 98 mV/cm for 25 hours; (2) then to an alternating electric field having a frequency of 90 MHz and a field strength of 98 mV/cm for 25 hours; (3) then to an alternating electric field having a frequency of 82 MHz and a field strength of 274 mV/cm for 32 hours; and (4) finally to an alternating electric field having a frequency of 90 MHz and a field strength of 274 mV/cm for 32 hours.

To test the ability of the EMF-treated IFFI1413 cells to degrade trichloromethane, industrial waste water was supplemented with trichloromethane to reconstitute a solution containing the chemical at 100 mg/L. 0.1 ml of the EMF-treated IFFI11413 cells at a concentration higher than 108 cells/ml was added to 100 L of the trichloromethane solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the trichloromethane solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 24 hours of incubation, the trichloromethane solutions were examined using gas chromatography. The results showed that after 24 hours of incubation, the trichloromethane concentration of solution A decreased more than 29% relative to solution C. In contrast, the trichloromethane concentration of solution B showed no significant change relative to solution C.

Example 21: Degradation of Toluene

15

20

25

30

Saccharomyces cerevisiae Hansen IFFI1399 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 76 MHz and a field strength of 89 mV/cm for 20 hours; (2) then to an alternating electric field having a frequency of 80 MHz and a field strength of 89 mV/cm for 20 hours; (3) then to an alternating electric field having a frequency of 86 MHz and

a field strength of 89 mV/cm for 20 hours; and (4) finally to an alternating electric field having a frequency of 96 MHz and a field strength of 89 mV/cm for 20 hours.

To test the ability of the EMF-treated IFFI1399 cells to degrade toluene, industrial waste water was supplemented with toluene to reconstitute a solution containing the chemical at 100 mg/L. 0.1 ml of the EMF-treated IFFI11399 cells at a concentration higher than 108 cells/ml was added to 100 L of the toluene solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the toluene solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 24 hours of incubation, the toluene solutions were examined using gas chromatography or HPLC. The results showed that after 24 hours of incubation, the toluene concentration of solution A decreased more than 32% relative to solution C. In contrast, the toluene concentration of solution B showed no significant change relative to solution C.

Example 22: Degradation of p-Xylene

15

20

Saccharomyces cerevisiae Hansen IFFI1336 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 72 MHz and a field strength of 93 mV/cm for 20 hours; (2) then to an alternating electric field having a frequency of 80 MHz and a field strength of 93 mV/cm for 20 hours; (3) then to an alternating electric field having a frequency of 88 MHz and a field strength of 93 mV/cm for 20 hours; and (4) finally to an alternating electric field having a frequency of 98 MHz and a field strength of 93 mV/cm for 20 hours.

To test the ability of the EMF-treated IFFI1336 cells to degrade pxylene, industrial waste water was supplemented with p-xylene to reconstitute a
solution containing the chemical at 100 mg/L. 0.1 ml of the EMF-treated
IFFI11336 cells at a concentration higher than 10⁸ cells/ml was added to 100 L of
the xylene solution and cultured at 28°C for 24 hours (solution A). One hundred
liters of the xylene solution containing the same number of non-treated cells
(solution B) or containing no cells (solution C) were used as controls. After 24
hours of incubation, the xylene solutions were examined using gas chromatography
or HPLC. The results showed that after 24 hours of incubation, the xylene

concentration of solution A decreased more than 24% relative to solution C. In contrast, the xylene concentration of solution B showed no significant change relative to solution C.

Example 23: Degradation of Benzaldehyde

5

10

15

20

25

Saccharomyces cerevisiae Hansen IFFI1331 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 78 MHz and a field strength of 130 mV/cm for 30 hours; (2) then to an alternating electric field having a frequency of 86 MHz and a field strength of 130 mV/cm for 30 hours; (3) then to an alternating electric field having a frequency of 94 MHz and a field strength of 130 mV/cm for 30 hours; and (4) finally to an alternating electric field having a frequency of 96 MHz and a field strength of 130 mV/cm for 30 hours.

benzaldehyde, industrial waste water was supplemented with benzaldehyde to reconstitute a solution containing the chemical at 100 mg/L. 0.1 ml of the EMF-treated IFFI11331 cells at a concentration higher than 10⁸ cells/ml was added to 100 L of the benzaldehyde solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the benzaldehyde solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 24 hours of incubation, the benzaldehyde solutions were examined using gas chromatography or HPLC. The results showed that after 24 hours of incubation, the benzaldehyde concentration of solution A decreased more than 34% relative to solution C. In contrast, the benzaldehyde concentration of solution B showed no significant change relative to solution C.

Example 24: Degradation of Propylaldehyde

Saccharomyces cerevisiae Hansen IFFI1396 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 76 MHz and a field strength of 103 mV/cm for 20 hours; (2) then to an alternating electric field having a frequency of 88 MHz and a field strength of 103 mV/cm for 20 hours; (3) then to an alternating electric field having a frequency of 96 MHz and

a field strength of 103 mV/cm for 20 hours; and (4) finally to an alternating electric field having a frequency of 98 MHz and a field strength of 103 mV/cm for 30 hours.

To test the ability of the EMF-treated IFFI1396 cells to degrade propylaldehyde, industrial waste water was supplemented with propylaldehyde to reconstitute a solution containing the chemical at 100 mg/L. 0.1 ml of the EMF-treated IFFI11396 cells at a concentration higher than 108 cells/ml was added to 100 L of the propylaldehyde solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the propylaldehyde solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 24 hours of incubation, the propylaldehyde solutions were examined using gas chromatography or HPLC. The results showed that after 24 hours of incubation, the propylaldehyde concentration of solution A decreased more than 19% relative to solution C. In contrast, the propylaldehyde concentration of solution B showed no significant change relative to solution C.

Example 25: Degradation of Enanthaldehyde

15

20

25

30

Saccharomyces cerevisiae Hansen IFFI1310 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 74 MHz and a field strength of 126 mV/cm for 40 hours; (2) then to an alternating electric field having a frequency of 82 MHz and a field strength of 126 mV/cm for 20 hours; (3) then to an alternating electric field having a frequency of 90 MHz and a field strength of 126 mV/cm for 30 hours; and (4) finally to an alternating electric field having a frequency of 98 MHz and a field strength of 126 mV/cm for 40 hours.

To test the ability of the EMF-treated IFFI1310 cells to degrade enanthaldehyde, industrial waste water was supplemented with enanthaldehyde to reconstitute a solution containing the chemical at 100 mg/L. 0.1 ml of the EMF-treated IFFI1310 cells at a concentration higher than 10⁸ cells/ml was added to 100 L of the enanthaldehyde solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the enanthaldehyde solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as

controls. After 24 hours of incubation, the enanthaldehyde solutions were examined using gas chromatography or HPLC. The results showed that after 24 hours of incubation, the enanthaldehyde concentration of solution A decreased more than 22% relative to solution C. In contrast, the enanthaldehyde concentration of solution B showed no significant change relative to solution C.

Example 26: Degradation of m-Dichlorobenzene

15

30

Saccharomyces cerevisiae Hansen IFFI1331 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 72 MHz and a field strength of 107 mV/cm for 20 hours; (2) then to an alternating electric field having a frequency of 80 MHz and a field strength of 107 mV/cm for 10 hours; (3) then to an alternating electric field having a frequency of 90 MHz and a field strength of 107 mV/cm for 30 hours; and (4) finally to an alternating electric field having a frequency of 94 MHz and a field strength of 107 mV/cm for 40 hours.

To test the ability of the EMF-treated IFFI1331 cells to degrade m-dichlorobenzene, waste water was supplemented with m-dichlorobenzene to reconstitute a solution containing the chemical at 100 mg/L. 0.1 ml of the EMF-treated IFFI1331 cells at a concentration higher than 108 cells/ml was added to 100 L of the m-dichlorobenzene solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the m-dichlorobenzene solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 24 hours of incubation, the m-dichlorobenzene solutions were examined using gas chromatography or HPLC. The results showed that after 24 hours of incubation, the m-dichlorobenzene concentration of solution A decreased more than 26% relative to solution C. In contrast, the m-dichlorobenzene concentration of solution B showed no significant change relative to solution C.

Example 27: Degradation of Acetophenone

Saccharomyces cerevisiae Hansen IFFI1311 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 76

MHz and a field strength of 124 mV/cm for 20 hours; (2) then to an alternating electric field having a frequency of 82 MHz and a field strength of 124 mV/cm for 30 hours; (3) then to an alternating electric field having a frequency of 90 MHz and a field strength of 124 mV/cm for 40 hours; and (4) finally to an alternating electric field having a frequency of 98 MHz and a field strength of 124 mV/cm for 20 hours.

To test the ability of the EMF-treated IFFI1311 cells to degrade acetophenone, waste water was supplemented with acetophenone to reconstitute a solution containing the chemical at 100 mg/L. 0.1 ml of the EMF-treated IFFI1311 cells at a concentration higher than 10⁸ cells/ml was added to 100 L of the acetophenone solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the acetophenone solution containing the same number of nontreated cells (solution B) or containing no cells (solution C) were used as controls. After 24 hours of incubation, the acetophenone solutions were examined using gas chromatography or HPLC. The results showed that after 24 hours of incubation, the acetophenone concentration of solution A decreased more than 11% relative to solution C. In contrast, the acetophenone concentration of solution B showed no significant change relative to solution C.

Example 28: Degradation of Arsanilic Acid

20

25

Saccharomyces cerevisiae Hansen IFFI1336 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 78 MHz and a field strength of 133 mV/cm for 30 hours; (2) then to an alternating electric field having a frequency of 88 MHz and a field strength of 133 mV/cm for 40 hours; (3) then to an alternating electric field having a frequency of 92 MHz and a field strength of 133 mV/cm for 30 hours; and (4) finally to an alternating electric field having a frequency of 96 MHz and a field strength of 133 mV/cm for 30 hours.

To test the ability of the EMF-treated IFFI1336 cells to degrade

arsanilic acid, waste water was supplemented with arsanilic acid to reconstitute a

solution containing the chemical at 100 mg/L. 0.1 ml of the EMF-treated IFFI1336

cells at a concentration higher than 108 cells/ml was added to 100 L of the arsanilic

acid solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the arsanilic acid solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 24 hours of incubation, the arsanilic acid solutions were examined using gas chromatography or HPLC. The results showed that after 24 hours of incubation, the arsanilic acid concentration of solution A decreased more than 13% relative to solution C. In contrast, the arsanilic acid concentration of solution B showed no significant change relative to solution C.

Example 29: Degradation of Roxarsone

10

15

20

Saccharomyces cerevisiae Hansen IFFI1338 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 78 MHz and a field strength of 110 mV/cm for 10 hours; (2) then to an alternating electric field having a frequency of 92 MHz and a field strength of 110 mV/cm for 10 hours; (3) then to an alternating electric field having a frequency of 78 MHz and a field strength of 213 mV/cm for 30 hours; and (4) finally to an alternating electric field having a frequency of 92 MHz and a field strength of 213 mV/cm for 30 hours.

To test the ability of the EMF-treated IFFI1338 cells to degrade roxarsone, waste water was supplemented with roxarsone to reconstitute a solution containing the chemical at 100 mg/L. 0.1 ml of the EMF-treated IFFI1338 cells at a concentration higher than 10⁸ cells/ml was added to 100 L of the roxarsone solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the roxarsone solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 24 hours of incubation, the roxarsone solutions were examined using gas chromatography or HPLC. The results showed that after 24 hours of incubation, the roxarsone concentration of solution A decreased more than 13% relative to solution C. In contrast, the roxarsone concentration of solution B showed no significant change relative to solution C.

Example 30: Degradation of Furazolidonum

Saccharomyces cerevisiae Hansen IFFI1413 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 74 MHz and a field strength of 98 mV/cm for 30 hours; (2) then to an alternating electric field having a frequency of 76 MHz and a field strength of 98 mV/cm for 20 hours; (3) then to an alternating electric field having a frequency of 86 MHz and a field strength of 98 mV/cm for 30 hours; and (4) finally to an alternating electric field having a frequency of 94 MHz and a field strength of 98 mV/cm for 30 hours.

To test the ability of the EMF-treated IFFI1413 cells to degrade furazolidonum, waste water was supplemented with furazolidonum to reconstitute a solution containing the chemical at 100 mg/L. 0.1 ml of the EMF-treated IFFI1413 cells at a concentration higher than 108 cells/ml was added to 100 L of the furazolidonum solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the furazolidonum solution containing the same number of nontreated cells (solution B) or containing no cells (solution C) were used as controls. After 24 hours of incubation, the furazolidonum solutions were examined using gas chromatography or HPLC. The results showed that after 24 hours of incubation, the furazolidonum concentration of solution A decreased more than 10% relative to solution C. In contrast, the furazolidonum concentration of solution B showed no significant change relative to solution C.

Example 31: Degradation of Decoquinate

10

20

30

Saccharomyces cerevisiae Hansen IFFI1411 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 78 MHz and a field strength of 112 mV/cm for 30 hours; (2) then to an alternating electric field having a frequency of 82 MHz and a field strength of 112 mV/cm for 30 hours; (3) then to an alternating electric field having a frequency of 86 MHz and a field strength of 112 mV/cm for 30 hours; and (4) finally to an alternating electric field having a frequency of 94 MHz and a field strength of 112 mV/cm for 20 hours.

To test the ability of the EMF-treated IFFI1411 cells to degrade decoquinate, waste water was supplemented with decoquinate to reconstitute a

solution containing the chemical at 100 mg/L. 0.1 ml of the EMF-treated IFFI1411 cells at a concentration higher than 10⁸ cells/ml was added to 100 L of the decoquinate solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the decoquinate solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 24 hours of incubation, the decoquinate solutions were examined using gas chromatography or HPLC. The results showed that after 24 hours of incubation, the decoquinate concentration of solution A decreased more than 17% relative to solution C. In contrast, the decoquinate concentration of solution B showed no significant change relative to solution C.

Example 32: Degradation of Trichlorphonum

10

Saccharomyces cerevisiae Hansen IFFI1211 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 74 MHz and a field strength of 219 mV/cm for 30 hours; (2) then to an alternating electric field having a frequency of 86 MHz and a field strength of 219 mV/cm for 20 hours; (3) then to an alternating electric field having a frequency of 96 MHz and a field strength of 219 mV/cm for 30 hours; and (4) finally to an alternating electric field having a frequency of 98 MHz and a field strength of 219 mV/cm for 20 hours.

To test the ability of the EMF-treated IFFI1211 cells to degrade trichlorphonum, waste water was supplemented with trichlorphonum to reconstitute a solution containing the chemical at 100 mg/L. 0.1 ml of the EMF-treated IFFI1211 cells at a concentration higher than 108 cells/ml was added to 100 L of the trichlorphonum solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the trichlorphonum solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 24 hours of incubation, the trichlorphonum solutions were examined using gas chromatography or HPLC. The results showed that after 24 hours of incubation, the trichlorphonum concentration of solution A decreased more than 15% relative to solution C. In contrast, the trichlorphonum concentration of solution B showed no significant change relative to solution C.

Example 33: Degradation of Dinitomidum and Zoalene

Saccharomyces cerevisiae Hansen IFFI1063 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 76 MHz and a field strength of 202 mV/cm for 30 hours; (2) then to an alternating electric field having a frequency of 82 MHz and a field strength of 202 mV/cm for 30 hours; (3) then to an alternating electric field having a frequency of 90 MHz and a field strength of 202 mV/cm for 20 hours; and (4) finally to an alternating electric field having a frequency of 96 MHz and a field strength of 202 mV/cm for 20 hours.

To test the ability of the EMF-treated IFFI1063 cells to degrade dinitomidum, waste water was supplemented with dinitomidum to reconstitute a solution containing the chemical at 100 mg/L. 0.1 ml of the EMF-treated IFFI1063 cells at a concentration higher than 108 cells/ml was added to 100 L of the dinitomidum solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the dinitomidum solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 24 hours of incubation, the dinitomidum solutions were examined using gas chromatography or HPLC. The results showed that after 24 hours of incubation, the dinitomidum concentration of solution A decreased more than 26% relative to solution C. In contrast, the dinitomidum concentration of solution B showed no significant change relative to solution C. The same method can be used to assess the zoalene-degrading ability of activated yeast cells.

Example 34: Degradation of Dodecane

20

25

Saccharomyces cerevisiae Hansen IFFI1213 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 72 MHz and a field strength of 168 mV/cm for 30 hours; (2) then to an alternating electric field having a frequency of 80 MHz and a field strength of 168 mV/cm for 20 hours; (3) then to an alternating electric field having a frequency of 90 MHz and a field strength of 168 mV/cm for 30 hours; and (4) finally to an alternating electric

field having a frequency of 98 MHz and a field strength of 168 mV/cm for 30 hours.

To test the ability of the EMF-treated IFFI1213 cells to degrade dodecane, waste water was supplemented with dodecane to reconstitute a solution containing the chemical at 100 mg/L. 0.1 ml of the EMF-treated IFFI1213 cells at a concentration higher than 10⁸ cells/ml was added to 100 L of the dodecane solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the dodecane solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 24 hours of incubation, the dodecane solutions were examined using gas chromatography or HPLC. The results showed that after 24 hours of incubation, the dodecane concentration of solution A decreased more than 21% relative to solution C. In contrast, the dodecane concentration of solution B showed no significant change relative to solution C.

Example 35: Degradation of Nonadecane

15

20

25

Saccharomyces cerevisiae Hansen IFFI1270 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 80 MHz and a field strength of 138 mV/cm for 30 hours; (2) then to an alternating electric field having a frequency of 82 MHz and a field strength of 138 mV/cm for 20 hours; (3) then to an alternating electric field having a frequency of 94 MHz and a field strength of 138 mV/cm for 24 hours; and (4) finally to an alternating electric field having a frequency of 98 MHz and a field strength of 138 mV/cm for 30 hours.

To test the ability of the EMF-treated IFFI1270 cells to degrade nonadecane, waste water was supplemented with nonadecane to reconstitute a solution containing the chemical at 100 mg/L. 0.1 ml of the EMF-treated IFFI1270 cells at a concentration higher than 10⁸ cells/ml was added to 100 L of the nonadecane solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the nonadecane solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 24 hours of incubation, the nonadecane solutions were examined using gas

chromatography or HPLC. The results showed that after 24 hours of incubation, the nonadecane concentration of solution A decreased more than 23% relative to solution C. In contrast, the nonadecane concentration of solution B showed no significant change relative to solution C:

Example 36: Degradation of Octacosane

5

15

20

25

Saccharomyces cerevisiae Hansen IFFI1293 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 76 MHz and a field strength of 138 mV/cm for 24 hours; (2) then to an alternating electric field having a frequency of 84 MHz and a field strength of 138 mV/cm for 24 hours; (3) then to an alternating electric field having a frequency of 96 MHz and a field strength of 138 mV/cm for 20 hours; and (4) finally to an alternating electric field having a frequency of 98 MHz and a field strength of 138 mV/cm for 40 hours.

To test the ability of the EMF-treated IFFI1293 cells to degrade octacosane, waste water was supplemented with octacosane to reconstitute a solution containing the chemical at 100 mg/L. 0.1 ml of the EMF-treated IFFI1293 cells at a concentration higher than 10⁸ cells/ml was added to 100 L of the octacosane solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the octacosane solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 24 hours of incubation, the octacosane solutions were examined using gas chromatography or HPLC. The results showed that after 24 hours of incubation, the octacosane concentration of solution A decreased more than 20% relative to solution C. In contrast, the octacosane concentration of solution B showed no significant change relative to solution C.

Example 37: Conversion of NH₄⁺

Saccharomyces cerevisiae Hansen AS2.614 were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 662 MHz and a field strength of 152 mV/cm for 18 hours; (2) then to an alternating electric field having a frequency of 666 MHz and a field strength of 152 mV/cm for

18 hours; (3) then to an alternating electric field having a frequency of 672 MHz and a field strength of 152 mV/cm for 18 hours; (4) then to an alternating electric field having a frequency of 678 MHz and a field strength of 152 mV/cm for 18 hours; (5) then to an alternating electric field having a frequency of 662 MHz and a field strength of 310 mV/cm for 25 hours; (6) then to an alternating electric field having a frequency of 666 MHz and a field strength of 310 mV/cm for 25 hours; (7) then to an alternating electric field having a frequency of 672 MHz and a field strength of 310 mV/cm for 35 hours; and (8) finally to an alternating electric field having a frequency of 678 MHz and a field strength of 310 mV/cm for 35 hours.

To test the ability of the EMF-treated AS2.614 cells to convert extracellular chemicals containing the NH₄⁺ ion into the cells' own biomass, waste water was supplemented with ammonium sulfate to reconstitute a solution containing ammonium sulfate at 200 mg/L. 0.1 ml of the EMF-treated AS2.614 cells at a concentration higher than 108 cells/ml was added to 100 L of the ammonium sulfate solution and cultured at 28°C for 48 hours (solution A). One hundred liters of the ammonium sulfate solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 48 hours of incubation, the ammonium sulfate solutions were examined using Kieldahl nitrogen-fixation test. The results showed that after 48 20 hours of incubation, the ammonium sulfate concentration of solution A decreased more than 22% relative to solution C. In contrast, the ammonium sulfate concentration of solution B showed no significant change relative to solution C.

Example 38: Conversion of Nitrates and Nitrites

10

25

Saccharomyces cerevisiae Hansen AS2.982 were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 661 MHz and a field strength of 126 mV/cm for 25 hours; (2) then to an alternating electric field having a frequency of 665 MHz and a field strength of 126 mV/cm for 25 hours; (3) then to an alternating electric field having a frequency of 672 MHz and a field strength of 126 mV/cm for 25 hours; (4) then to an alternating electric field having a frequency of 676 MHz and a field strength of 126 mV/cm for 25 hours; (5) then to an alternating electric field having a frequency of 661 MHz and a

field strength of 196 mV/cm for 25 hours; (6) then to an alternating electric field having a frequency of 665 MHz and a field strength of 196 mV/cm for 25 hours; (7) then to an alternating electric field having a frequency of 672 MHz and a field strength of 196 mV/cm for 38 hours; and (8) finally to an alternating electric field having a frequency of 676 MHz and a field strength of 196 mV/cm for 38 hours.

To test the ability of the EMF-treated AS2.982 cells to convert extracellular nitrates and/or nitrites into the cells' own biomass, waste water was supplemented with sodium nitrate and sodium nitrite to reconstitute a solution containing sodium nitrate/sodium nitrite at a total concentration of 200 mg/L. 0.1 ml of the EMF-treated AS2.982 cells at a concentration higher than 108 cells/ml was added to 100 L of the sodium nitrate/sodium nitrite solution and cultured at 28°C for 48 hours (solution A). One hundred liters of the nitrate/nitrite solution containing the same number of non-treated cells (solution B) or containing no cells (solution C) were used as controls. After 48 hours of incubation, the nitrate/nitrite solutions were examined using Kjeldahl nitrogen-fixation test. The results showed that after 48 hours of incubation, the nitrate/nitrite concentration of solution A decreased more than 29% relative to solution C. In contrast, the nitrate/nitrite concentration of solution B showed no significant change relative to solution C.

Example 39: Conversion of PO₄³, HPO₄², H₂PO₄, and/or H₃PO₄ in a Culture Medium Into Intracellular Phosphorus

20

Saccharomyces cerevisiae Hansen AS2.620 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 98 MHz and a field strength of 68 mV/cm for 24 hours; (2) then to an alternating electric field having a frequency of 112 MHz and a field strength of 68 mV/cm for 24 hours; (3) then to an alternating electric field having a frequency of 108 MHz and a field strength of 68 mV/cm for 24 hours; (4) then to an alternating electric field having a frequency of 118 MHz and a field strength of 68 mV/cm for 24 hours; (5) then to an alternating electric field having a frequency of 98 MHz and a field strength of 240 mV/cm for 24 hours; (6) then to an alternating electric field having a frequency of 112 MHz and a field strength of 240 mV/cm for 24 hours; (7) then to an alternating electric field having a frequency of 108 MHz and a field

strength of 240 mV/cm for 42 hours; and (8) finally to an alternating electric field having a frequency of 118 MHz and a field strength of 240 mV/cm for 42 hours.

To test the ability of the EMF-treated AS2.620 cells to convert biologically available phosphorus to intracellular phosphorus, waste water or filtrate from animal manure or garbage was supplemented with Na₃PO₄ to reconstitute a solution containing Na₃PO₄ at 200 mg/L. 0.1 ml of the EMF-treated AS2.620 cells at a concentration higher than 10⁸ cells/ml was added to 100 L of the Na₃PO₄ solution and cultured at 28°C for 48 hours (solution A). One hundred liters of the Na₃PO₄ solution containing the same number of non-treated yeast cells (solution B) or containing no yeast cells (solution C) were used as controls. After 48 hours of incubation, the solutions were examined using ultraviolet spectrophotometry. The results showed that after 48 hours of incubation, the Na₃PO₄ concentration in solution A decreased more than 23% relative to solution C. In contrast, the Na₃PO₄ concentration in solution B had no significant change relative to solution C.

Example 40: Reduction of Odor Caused by Hydrogen Sulfide

15

20

25

Saccharomyces cerevisiae Hansen Var. ellipsoideus (Hansen)

Dekker AS2.559 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 2165 MHz and a field strength of 240 mV/cm for 20 hours; (2) then to an alternating electric field having a frequency of 2175 MHz and a field strength of 240 mV/cm for 20 hours; (3) then to an alternating electric field having a frequency of 2200 MHz and a field strength of 240 mV/cm for 20 hours; and (4) finally to an alternating electric field having a frequency of 2235 MHz and a field strength of 240 mV/cm for 20 hours.

To test the ability of the EMF-treated AS2.559 cells to reduce odor caused by hydrogen sulfide, waste water or filtrate from animal manure or garbage was supplemented with H₂S to reconstitute a solution containing H₂S at 100 mg/L. 0.1 ml of the EMF-treated AS2.559 cells at a concentration higher than 10⁸ cells/ml was added to 100 L of the H₂S solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the H₂S solution containing the same number of non-treated yeast cells (solution B) or containing no yeast cells (solution C) were used

as controls. After 24 hours of incubation, the solutions were examined using mass spectrometry (MAS-nose, manufactured by VG). The results showed that after 24 hours of incubation, the H₂S concentration of solution A decreased more than 13% relative to solution C. In contrast, the H₂S concentration of solution B had no significant change relative to solution C.

Example 41: Reduction of Odor Caused by Ammonia

15

Saccharomyces cerevisiae Hansen AS2.423 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 2160 MHz and a field strength of 250 mV/cm for 20 hours; (2) then to an alternating electric field having a frequency of 2175 MHz and a field strength of 250 mV/cm for 20 hours; (3) then to an alternating electric field having a frequency of 2210 MHz and a field strength of 250 mV/cm for 20 hours; and (4) finally to an alternating electric field having a frequency of 2245 MHz and a field strength of 250 mV/cm for 10 hours.

To test the ability of the EMF-treated AS2.423 cells to reduce odor caused by ammonia, waste water or filtrate from animal manure or garbage was supplemented with ammonia to reconstitute a solution containing ammonia (in the form of ammonium hydroxide) at 100 mg/L. 0.1 ml of the EMF-treated AS2.423 cells at a concentration higher than 10⁸ cells/ml was added to 100 L of the ammonia solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the ammonia solution containing the same number of non-treated yeast cells (solution B) or containing no yeast cells (solution C) were used as controls. After 24 hours of incubation, the solutions were examined using mass spectrometry (MAS-nose, manufactured by VG). The results showed that after 24 hours of incubation, the ammonia concentration of solution A decreased more than 11% relative to solution C. In contrast, the ammonia concentration of solution B had no significant change relative to solution C.

Example 42: Reduction of Odor Caused by Indole

30 Saccharomyces cerevisiae Hansen Var. ellipsoideus (Hansen)

Dekker AS2.612 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an

PCT/GB01/05439 WO 02/070682

alternating electric field having a frequency of 2165 MHz and a field strength of 240 mV/cm for 40 hours; (2) then to an alternating electric field having a frequency of 2180 MHz and a field strength of 240 mV/cm for 20 hours; (3) then to an alternating electric field having a frequency of 2200 MHz and a field strength of 240 mV/cm for 40 hours; and (4) finally to an alternating electric field having a frequency of 2220 MHz and a field strength of 240 mV/cm for 20 hours.

To test the ability of the EMF-treated AS2.612 cells to reduce odor caused by indole, waste water or filtrate from animal manure or garbage was supplemented with indole to reconstitute a solution containing indole at 100 mg/L. 10 0.1 ml of the EMF-treated AS2.612 cells at a concentration higher than 108 cells/ml was added to 100 L of the indole solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the indole solution containing the same number of non-treated yeast cells (solution B) or containing no yeast cells (solution C) were used as controls. After 24 hours of incubation, the solutions were examined using mass spectrometry (MAS-nose, manufactured by VG). The results showed that after 24 hours of incubation, the indole concentration of solution A decreased more than 15% relative to solution C. In contrast, the indole concentration of solution B had no significant change relative to solution C.

15

20

30

Example 43: Reduction of Odor Caused by Methylamine, Dimethylamine or Trimethylamine

Saccharomyces cerevisiae Hansen Var. ellipsoideus (Hansen) Dekker AS2.541 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 2160 MHz and a field strength of 250 mV/cm for 20 hours; (2) then to an alternating electric field having a frequency of 2190 MHz and a field strength of 250 mV/cm for 10 hours; (3) then to an alternating electric field having a frequency of 2210 MHz and a field strength of 250 mV/cm for 40 hours; and (4) finally to an alternating electric field having a frequency of 2250 MHz and a field strength of 250 mV/cm for 40 hours.

To test the ability of the EMF-treated AS2.541 cells to reduce odor caused by methylamine, dimethylamine or trimethylamine, waste water or filtrate from animal manure or garbage was supplemented with methylamine,

dimethylamine, or trimethylamine to reconstitute a solution containing methylamine, dimethylamine, or trimethylamine at 100 mg/L. 0.1 ml of the EMFtreated AS2.541 cells at a concentration higher than 108 cells/ml was added to 100 L of the methylamine, dimethylamine or trimethylamine solution and cultured at 5 28°C for 24 hours (solution A). One hundred liters of the methylamine. dimethylamine or trimethylamine solution containing the same number of nontreated yeast cells (solution B) or containing no yeast cells (solution C) were used as controls. After 24 hours of incubation, the solutions were examined using mass spectrometry (MAS-nose, manufactured by VG). The results showed that after 24 10 hours of incubation, the methylamine, dimethylamine or trimethylamine concentration of solution A decreased more than 23% relative to solution C. In contrast, the methylamine, dimethylamine, or trimethylamine concentration of solution B had no significant change relative to solution C.

Example 44: Reduction of Odor Caused by Organic Acids

15

25

Saccharomyces cerevisiae Hansen Var. ellipsoideus (Hansen) Dekker AS2.53 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 2315 MHz and a field strength of 290 mV/cm for 30 hours; (2) then to an alternating electric field having a frequency of 2335 MHz and a field strength of 290 mV/cm for 10 hours; (3) then to an alternating electric field having a frequency of 2355 MHz and a field strength of 290 mV/cm for 20 hours; and (4) finally to an alternating electric field having a frequency of 2375 MHz and a field strength of 290 mV/cm for 10 hours.

To test the ability of the EMF-treated AS2.53 cells to reduce odor caused by organic acids, waste water or filtrate from animal manure or garbage was supplemented with acetic acid to reconstitute a solution containing acetic acid at 100 mg/L. 0.1 ml of the EMF-treated AS2.53 cells at a concentration higher than 108 cells/ml was added to 100 L of the acetic acid solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the acetic acid solution containing the same number of non-treated yeast cells (solution B) or containing no yeast cells 30 (solution C) were used as controls. After 24 hours of incubation, the solutions were examined using mass spectrometry (MAS-nose, manufactured by VG). The

results showed that after 24 hours of incubation, the acetic acid concentration of solution A decreased more than 19% relative to solution C. In contrast, the acetic acid concentration of solution B had no significant change relative to solution C.

Example 45: Reduction of Odor Caused by p-Cresol

5

10

15

20

25

Saccharomyces cerevisiae Hansen Var. ellipsoideus (Hansen)

Dekker AS2.163 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 2300 MHz and a field strength of 98 mV/cm for 20 hours; (2) then to an alternating electric field having a frequency of 2370 MHz and a field strength of 98 mV/cm for 15 hours; (3) then to an alternating electric field having a frequency of 2300 MHz and a field strength of 250 mV/cm for 20 hours; and (4) finally to an alternating electric field having a frequency of 2370 MHz and a field strength of 250 mV/cm for 30 hours.

caused by p-cresol, waste water or filtrate from animal manure or garbage was supplemented with p-cresol to reconstitute a solution containing p-cresol at 100 mg/L. 0.1 ml of the EMF-treated AS2.163 cells at a concentration higher than 108 cells/ml was added to 100 L of the p-cresol solution and cultured at 28°C for 24 hours (solution A). One hundred liters of the p-cresol solution containing the same number of non-treated yeast cells (solution B) or containing no yeast cells (solution C) were used as controls. After 24 hours of incubation, the solutions were examined using mass spectrometry (MAS-nose, manufactured by VG). The results showed that after 24 hours of incubation, the p-cresol concentration of solution A decreased more than 23% relative to solution C. In contrast, the p-cresol concentration of solution B had no significant change relative to solution C.

Example 46: Suppression of the Growth of Staphylococcus aures

Saccharomyces cerevisiae Hansen IFFI1037 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 30 MHz and a field strength of 26 mV/cm for 12 hours; (2) then to an alternating electric field having a frequency of 36 MHz and a field strength of 26 mV/cm for 12 hours; (3) then to an alternating electric field having a frequency of 43 MHz and

a field strength of 26 mV/cm for 12 hours; (4) then to an alternating electric field having a frequency of 47 MHz and a field strength of 26 mV/cm for 12 hours; (5) then to an alternating electric field having a frequency of 30 MHz and a field strength of 150 mV/cm for 24 hours; (6) then to an alternating electric field having a frequency of 36 MHz and a field strength of 150 mV/cm for 24 hours; (7) then to an alternating electric field having a frequency of 43 MHz and a field strength of 150 mV/cm for 24 hours; and (8) finally to an alternating electric field having a frequency of 47 MHz and a field strength of 150 mV/cm for 24 hours.

To test the ability of the EMF-treated IFFI1037 cells to suppress the growth of Staphylococcus aures, waste water or filtrate from animal manure or garbage containing Staphylococcus aures was incubated under routine conditions to reconstitute a solution containing Staphylococcus aures at more than 10¹⁰ cells/ml. One milliliter of the EMF-treated IFFI1037 cells at a concentration of 2 x 10⁸ - 5 x 10⁸ cells/ml was added to 1 L of the Staphylococcus aures solution and cultured at 30°C for 24 hours (solution A). One liter of the Staphylococcus aures solution containing the same number of non-treated yeast cells (solution B) or containing no yeast cells (solution C) was used as controls. After 24 hours of incubation, the solutions were examined using a flow cytometer. The results showed that after 24 hours of incubation, the number of live Staphylococcus aures in solution A decreased more than 2.7% relative to solution C. In contrast, the number of live Staphylococcus aures in solution B showed no significant change relative to solution C.

10

20

25

Example 47: Suppression of the Growth of Diplococcus pneumonia

Saccharomyces cerevisiae Hansen IFFI1021 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 30 MHz and a field strength of 26 mV/cm for 12 hours; (2) then to an alternating electric field having a frequency of 36 MHz and a field strength of 26 mV/cm for 12 hours; (3) then to an alternating electric field having a frequency of 42 MHz and a field strength of 26 mV/cm for 12 hours; (4) then to an alternating electric field having a frequency of 49 MHz and a field strength of 26 mV/cm for 12 hours; (5) then to an alternating electric field having a frequency of 30 MHz and a field

5

20

strength of 150 mV/cm for 24 hours; (6) then to an alternating electric field having a frequency of 36 MHz and a field strength of 150 mV/cm for 24 hours; (7) then to an alternating electric field having a frequency of 42 MHz and a field strength of 150 mV/cm for 24 hours; and (8) finally to an alternating electric field having a frequency of 49 MHz and a field strength of 150 mV/cm for 24 hours.

To test the ability of the EMF-treated IFFI1021 cells to suppress the growth of *Diplococcus pneumonia*, waste water or filtrate from animal manure or garbage containing *Diplococcus pneumonia* was incubated under routine conditions to reconstitute a solution containing *Diplococcus pneumonia* at more than 10¹⁰

10 cells/ml. One milliliter of the EMF-treated IFFI1021 cells at a concentration of 2 x 10⁸ - 5 x 10⁸ cells/ml was added to 1 L of the *Diplococcus pneumonia* solution and cultured at 30°C for 24 hours (solution A). One liter of the *Diplococcus pneumonia* solution containing the same number of non-treated yeast cells (solution B) or containing no yeast cells (solution C) was used as controls. After 24 hours of incubation, the solutions were examined using a flow cytometer. The results showed that after 24 hours of incubation, the number of live *Diplococcus pneumonia* in solution A decreased more than 2.8% relative to solution C. In contrast, the number of live *Diplococcus pneumonia* in solution B showed no significant change relative to solution C.

Example 48: Suppression of the Growth of Bacillus anthracis

Saccharomyces cerevisiae Hansen IFFI1251 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 35 MHz and a field strength of 26 mV/cm for 12 hours; (2) then to an alternating electric field having a frequency of 39 MHz and a field strength of 26 mV/cm for 12 hours; (3) then to an alternating electric field having a frequency of 43 MHz and a field strength of 26 mV/cm for 12 hours; (4) then to an alternating electric field having a frequency of 47 MHz and a field strength of 26 mV/cm for 12 hours; (5) then to an alternating electric field having a frequency of 35 MHz and a field strength of 150 mV/cm for 24 hours; (6) then to an alternating electric field having a frequency of 39 MHz and a field strength of 150 mV/cm for 24 hours; (7) then to an alternating electric field having a frequency of 43 MHz and a field strength of

150 mV/cm for 24 hours; and (8) finally to an alternating electric field having a frequency of 47 MHz and a field strength of 150 mV/cm for 24 hours.

To test the ability of the EMF-treated IFFI1251 cells to suppress the growth of *Bacillus anthracis*, waste water or filtrate from animal manure or garbage containing *Bacillus anthracis* was incubated under routine conditions to reconstitute a solution containing *Bacillus anthracis* at more than 10^{10} cells/ml. One milliliter of the EMF-treated IFFI1251 cells at a concentration of 2 x 10^8 - 5 x 10^8 cells/ml was added to 1 L of the *Bacillus anthracis* solution and cultured at 30° C for 24 hours (solution A). One liter of the *Bacillus anthracis* solution containing the same number of non-treated yeast cells (solution B) or containing no yeast cells (solution C) was used as controls. After 24 hours of incubation, the solutions were examined using a flow cytometer. The results showed that after 24 hours of incubation, the number of live *Bacillus anthracis* in solution A decreased more than 3.1% relative to solution C. In contrast, the number of live *Bacillus anthracis* in solution C.

10

15

20

25

30

Example 49: Suppression of the Growth of Mycobacterium tuberculosis

Saccharomyces cerevisiae Hansen IFFI1331 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 33 MHz and a field strength of 26 mV/cm for 12 hours; (2) then to an alternating electric field having a frequency of 36 MHz and a field strength of 26 mV/cm for 12 hours; (3) then to an alternating electric field having a frequency of 45 MHz and a field strength of 26 mV/cm for 12 hours; (4) then to an alternating electric field having a frequency of 47 MHz and a field strength of 26 mV/cm for 12 hours; (5) then to an alternating electric field having a frequency of 33 MHz and a field strength of 150 mV/cm for 24 hours; (6) then to an alternating electric field having a frequency of 36 MHz and a field strength of 150 mV/cm for 24 hours; (7) then to an alternating electric field having a frequency of 45 MHz and a field strength of 150 mV/cm for 24 hours; and (8) finally to an alternating electric field having a frequency of 47 MHz and a field strength of 150 mV/cm for 24 hours.

To test the ability of the EMF-treated IFFI1331 cells to suppress the growth of *Mycobacterium tuberculosis*, waste water or filtrate from animal manure

or garbage containing *Mycobacterium tuberculosis* was incubated under routine conditions to reconstitute a solution containing *Mycobacterium tuberculosis* at more than 10¹⁰ cells/ml. One milliliter of the EMF-treated IFFI1331 cells at a concentration of 2 x 10⁸ - 5 x 10⁸ cells/ml was added to 1 L of the *Mycobacterium tuberculosis* solution and cultured at 30°C for 24 hours (solution A). One liter of the *Mycobacterium tuberculosis* solution containing the same number of nontreated yeast cells (solution B) or containing no yeast cells (solution C) was used as controls. After 24 hours of incubation, the solutions were examined using a flow cytometer. The results showed that after 24 hours of incubation, the number of live *Mycobacterium tuberculosis* in solution A decreased more than 2.9% relative to solution C. In contrast, the number of live *Mycobacterium tuberculosis* in solution B showed no significant change relative to solution C.

Example 50: Suppression of the Growth of E. Coli

15

Saccharomyces cerevisiae Hansen IFFI1345 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 30 MHz and a field strength of 26 mV/cm for 12 hours; (2) then to an alternating electric field having a frequency of 34 MHz and a field strength of 26 mV/cm for 12 hours; (3) then to an alternating electric field having a frequency of 38 MHz and a field strength of 26 mV/cm for 12 hours; (4) then to an alternating electric field having a frequency of 49 MHz and a field strength of 26 mV/cm for 12 hours; (5) then to an alternating electric field having a frequency of 30 MHz and a field strength of 150 mV/cm for 24 hours; (6) then to an alternating electric field having a frequency of 34 MHz and a field strength of 150 mV/cm for 24 hours; (7) then to an alternating electric field having a frequency of 38 MHz and a field strength of 150 mV/cm for 24 hours; and (8) finally to an alternating electric field having a frequency of 49 MHz and a field strength of 150 mV/cm for 24 hours.

To test the ability of the EMF-treated IFFI1345 cells to suppress the growth of E. Coli, waste water or filtrate from animal manure or garbage containing E. Coli was incubated under routine conditions to reconstitute a solution containing E. Coli at more than 10^{10} cells/ml. One milliliter of the EMF-treated IFFI1345 cells at a concentration of 2×10^8 - 5×10^8 cells/ml was added to 1 L of

the *E. Coli* solution and cultured at 30°C for 24 hours (solution A). One liter of the *E. Coli* solution containing the same number of non-treated yeast cells (solution B) or containing no yeast cells (solution C) was used as controls. After 24 hours of incubation, the solutions were examined using a flow cytometer. The results showed that after 24 hours of incubation, the number of live *E. Coli* in solution A decreased more than 48% relative to solution C. In contrast, the number of live *E. Coli* in solution B showed no significant change relative to solution C.

Example 51: Suppression of the Growth of Salmonella

the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 30 MHz and a field strength of 26 mV/cm for 12 hours; (2) then to an alternating electric field having a frequency of 33 MHz and a field strength of 26 mV/cm for 12 hours; (3) then to an alternating electric field having a frequency of 36 MHz and a field strength of 26 mV/cm for 12 hours; (4) then to an alternating electric field having a frequency of 38 MHz and a field strength of 26 mV/cm for 12 hours; (5) then to an alternating electric field having a frequency of 30 MHz and a field strength of 150 mV/cm for 24 hours; (6) then to an alternating electric field having a frequency of 33 MHz and a field strength of 150 mV/cm for 24 hours; (7) then to 20 an alternating electric field having a frequency of 36 MHz and a field strength of 150 mV/cm for 24 hours; and (8) finally to an alternating electric field having a frequency of 38 MHz and a field strength of 150 mV/cm for 24 hours; and (8) finally to an alternating electric field having a frequency of 38 MHz and a field strength of 150 mV/cm for 24 hours.

To test the ability of the EMF-treated IFFI1211 cells to suppress the growth of Salmonella, waste water or filtrate from animal manure or garbage containing Salmonella was incubated under routine conditions to reconstitute a solution containing Salmonella at more than 10¹⁰ cells/ml. One milliliter of the EMF-treated IFFI1211 cells at a concentration of 2 x 10⁸ - 5 x 10⁸ cells/ml was added to 1 L of the Salmonella solution and cultured at 30°C for 24 hours (solution A). One liter of the Salmonella solution containing the same number of non-treated yeast cells (solution B) or containing no yeast cells (solution C) was used as controls. After 24 hours of incubation, the solutions were examined using a flow cytometer. The results showed that after 24 hours of incubation, the number of live

Salmonella in solution A decreased more than 66% relative to solution C. In contrast, the number of live Salmonella in solution B showed no significant change relative to solution C.

Example 52: Suppression of the Growth of Green Algae

5

10

20

25

Saccharomyces cerevisiae Hansen AS2.408 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 6353 MHz and a field strength of 112 mV/cm for 24 hours; (2) then to an alternating electric field having a frequency of 6357 MHz and a field strength of 112 mV/cm for 24 hours; (3) then to an alternating electric field having a frequency of 6364 MHz and a field strength of 112 mV/cm for 24 hours; (4) then to an alternating electric field having a frequency of 6368 MHz and a field strength of 112 mV/cm for 24 hours; (5) then to an alternating electric field having a frequency of 6353 MHz and a field strength of 290 mV/cm for 56 hours; (6) then to an alternating electric field having a frequency of 6364 MHz and a field strength of 290 mV/cm for 24 hours; and (8) finally to an alternating electric field having a frequency of 6368 MHz and a field strength of 290 mV/cm for 24 hours; and (8) finally to an alternating electric field having a frequency of 6368 MHz and a field strength of 290 mV/cm for 24 hours.

To test the ability of the EMF-treated AS2.408 cells to suppress the growth of green algae, lake water or other surface water containing green algae was cultured under routine conditions to reconstitute a solution containing green algae at more than $1.0x10^9 - 1.5x10^9$ cells/ml. 0.1ml of the EMF-treated AS2.408 cells at a concentration of 10^8 cells/ml was added to 1 L of the green algae solution and cultured at $28-32^{\circ}$ C for 72 hours (solution A). One liter of the green algae solution containing the same number of non-treated yeast cells (solution B) or containing no yeast cells (solution C) were used as controls. After 72 hours of incubation, the solutions were examined using a flow cytometer. The results showed that after 72 hours of incubation, the number of live green algae in solution A decreased more than 28% relative to solution C. In contrast, the number of live green algae in solution B showed no significant change relative to solution C.

Example 53: Suppression of the Growth of Blue Algae

Saccharomyces cerevisiae Hansen AS2.414 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 6355 MHz and a field strength of 85 mV/cm for 24 hours; (2) then to an alternating electric field having a frequency of 6360 MHz and a field strength of 85 mV/cm for 24 hours; (3) then to an alternating electric field having a frequency of 6364 MHz and a field strength of 85 mV/cm for 24 hours; (4) then to an alternating electric field having a frequency of 6367 MHz and a field strength of 85 mV/cm for 24 hours; (5) then to an alternating electric field having a frequency of 6355 MHz and a field strength of 250 mV/cm for 56 hours; (6) then to an alternating electric field having a frequency of 6360 MHz and a field strength of 250 mV/cm for 56 hours; (7) then to an alternating electric field having a frequency of 6364 MHz and a field strength of 250 mV/cm for 24 hours; and (8) finally to an alternating electric field having a frequency of 6367 MHz and a field strength of 250 mV/cm for 24 hours.

To test the ability of the EMF-treated AS2.414 cells to suppress the growth of blue algae, lake water or other surface water containing blue algae was cultured under routine conditions to reconstitute a solution containing blue algae at more than $1.0x10^9 - 1.5x10^9$ cells/ml. 0.1ml of the EMF-treated AS2.414 cells at a concentration of 10^8 cells/ml was added to 1 L of the blue algae solution and cultured at $28-32^{\circ}$ C for 72 hours (solution A). One liter of the blue algae solution containing the same number of non-treated yeast cells (solution B) or containing no yeast cells (solution C) were used as controls. After 72 hours of incubation, the solutions were examined using a flow cytometer. The results showed that after 72 hours of incubation, the number of live blue algae in solution A decreased more than 31% relative to solution C. In contrast, the number of live blue algae in solution B showed no significant change relative to solution C.

Example 54: Suppression of the Growth of Red Algae

15

20

25

Saccharomyces cerevisiae Hansen AS2.416 cells were cultured in the presence of a series of alternating electric fields in the following sequence: the yeast cells were exposed to (1) an alternating electric field having a frequency of 6352 MHz and a field strength of 136 mV/cm for 24 hours; (2) then to an alternating electric field having a frequency of 6359 MHz and a field strength of

of 6363 MHz and a field strength of 136 mV/cm for 24 hours; (4) then to an alternating electric field having a frequency of 6370 MHz and a field strength of 136 mV/cm for 24 hours; (5) then to an alternating electric field having a frequency of 6370 MHz and a field strength of 136 mV/cm for 24 hours; (5) then to an alternating electric field having a frequency of 6352 MHz and a field strength of 337 mV/cm for 56 hours; (6) then to an alternating electric field having a frequency of 6359 MHz and a field strength of 337 mV/cm for 56 hours; (7) then to an alternating electric field having a frequency of 6363 MHz and a field strength of 337 mV/cm for 24 hours; and (8) finally to an alternating electric field having a frequency of 6370 MHz and a field strength of 337 mV/cm for 24 hours.

To test the ability of the EMF-treated AS2.416 cells to suppress the growth of red algae, lake water or other surface water containing red algae was cultured under routine conditions to reconstitute a solution containing red algae at more than $1.0x10^9 - 1.5x10^9$ cells/ml. 0.1ml of the EMF-treated AS2.416 cells at a concentration of 10^8 cells/ml was added to 1 L of the red algae solution and cultured at 28-32°C for 72 hours (solution A). One liter of the red algae solution containing the same number of non-treated yeast cells (solution B) or containing no yeast cells (solution C) were used as controls. After 72 hours of incubation, the solutions were examined using a flow cytometer. The results showed that after 72 hours of incubation, the number of live red algae in solution A decreased more than 37% relative to solution C. In contrast, the number of live red algae in solution B showed no significant change relative to solution C.

Example 55: Decomposition of Algal Debris

10

15

20

Saccharomyces cerevisiae Hansen AS2.422 cells were cultured in
the presence of a series of alternating electric fields in the following sequence: the
yeast cells were exposed to (1) an alternating electric field having a frequency of
4452 MHz and a field strength of 127 mV/cm for 32 hours; (2) then to an
alternating electric field having a frequency of 4456 MHz and a field strength of
127 mV/cm for 32 hours; (3) then to an alternating electric field having a frequency
of 4462 MHz and a field strength of 127 mV/cm for 32 hours; (4) then to an
alternating electric field having a frequency of 4464 MHz and a field strength of
127 mV/cm for 32 hours; (5) then to an alternating electric field having a frequency

of 4452 MHz and a field strength of 268 mV/cm for 32 hours; (6) then to an alternating electric field having a frequency of 4456 MHz and a field strength of 268 mV/cm for 32 hours; (7) then to an alternating electric field having a frequency of 4462 MHz and a field strength of 268 mV/cm for 64 hours; and (8) finally to an alternating electric field having a frequency of 4464 MHz and a field strength of 268 mV/cm for 64 hours.

To test the ability of the EMF-treated AS2.422 cells to decompose debris of algae, lake water or other surface water containing debris of green, blue and/or red algae was cultured under routine conditions to reconstitute a solution containing debris of green, blue and/or red algae at more than $1.0 \times 10^9 - 1.5 \times 10^9$ cells/ml. 0.1ml of the EMF-treated AS2.422 cells at a concentration of 10^8 cells/ml was added to 1 L of the algae solution and cultured at $28-32^{\circ}$ C for 72 hours (solution A). One liter of the algae solution containing the same number of nontreated yeast cells (solution B) or containing no yeast cells (solution C) were used as controls. After 72 hours of incubation, the solutions were examined. The results showed that after 72 hours of incubation, the number of green, blue and/or red algae in solution A decreased more than 26% relative to solution C. In contrast, the number of green, blue and/or red algae in solution B showed no significant change relative to solution C.

While a number of embodiments of this invention have been set forth, it is apparent that the basic constructions may be altered to provide other embodiments which utilize the compositions and methods of this invention.

CLAIMS

What is claimed is:

15

20

1. A composition comprising one or more of the following pluralities

of yeast cells:

- 1) first plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to degrade a polymeric compound in a culture medium as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 4230 to 4260 MHz and a field strength in the range of 0.5 to 360 mV/cm, as compared to yeast cells not having been so cultured;
- 2) second plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to degrade a nitrogen-containing compound in a culture medium as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 5520 to 5540 MHz and a field strength in the range of 0.5 to 360 mV/cm, as compared to yeast cells not having been so cultured;
- 3) third plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to degrade an antibiotic or an organic solvent in a culture medium as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 70 to 100 MHz and a field strength in the range of 0.5 to 350 mV/cm, as compared to yeast cells not having been so cultured;
- 4) fourth plurality of yeast cells, wherein said plurality of yeast cells are
 25 characterized by a substantial increase in their capability to convert bio-available
 nitrogen in a culture medium into intracellular nitrogen as a result of having been
 cultured in the presence of an alternating electric field having a frequency in the
 range of 660 to 680 MHz and a field strength in the range of 0.1 to 350 mV/cm, as
 compared to yeast cells not having been so cultured;
- 5) fifth plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to convert biologically

5

10

15

20

available phosphorus in a culture medium into intracellular phosphorus as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 80 to 440 MHz and a field strength in the range of 0.5 to 350 mV/cm, as compared to yeast cells not having been so cultured;

- 6) sixth plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to reduce odor of a culture medium as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 2160 to 2380 MHz and a field strength in the range of 0.5 to 320 mV/cm, as compared to yeast cells not having been so cultured;
- 7) seventh plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to suppress the growth of pathogenic microbes as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 30 to 50 MHz and a field strength in the range of 0.5 to 200 mV/cm, as compared to yeast cells not having been so cultured; and
- 8) eighth plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to decompose debris of algae as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 4440 to 4470 MHz and a field strength in the range of 0.5 to 400 mV/cm, as compared to yeast cells not having been so cultured.
- The composition of claim 1 comprising first plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in
 their capability to degrade a polymeric compound in a culture medium as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 4230 to 4260 MHz and a field strength in the range of 0.5 to 360 mV/cm, as compared to yeast cells not having been so cultured.
- 3. The composition of claim 2, wherein said frequency is in the range of 4240 to 4260 MHz.

5

- 4. The composition of claim 2, wherein said field strength is in the range of 50 to 360 mV/cm.
- 5. The composition of claim 2, wherein said yeast cells are derived from cells of the species Saccharomyces cerevisiae, Saccharomyces carlsbergensis, or Hansenula subpelliculosa.
- The composition of claim 2, wherein said yeast cells are derived from cells of the strain deposited at the China General Microbiological Culture Collection Center with an accession number selected from the group consisting of AS2.11, AS2.53, AS2.56, AS2.70, AS2.98, AS2.101, AS2.168, AS2.374,
 AS2.406, AS2.409, AS2.430, AS2.453, AS2.463, AS2.467, AS2.502, AS2.516, AS2.536, AS2.541, AS2.443, AS2.459, AS2.738, AS2.740, and IFFI1331.
 - 7. The composition of claim 2, wherein said polymeric compound is cellulose.
- 8. The composition of claim 7, wherein said frequency is in the range of 4240-4260 MHz, and said field strength is in the range of 80-300 mV/cm.
 - 9. The composition of claim 2, wherein said polymeric compound is hemicellulose.
 - 10. The composition of claim 9, wherein said frequency is in the range of 4240-4260 MHz, and said field strength is in the range of 60-270 mV/cm.
- 20 11. The composition of claim 2, wherein said polymeric compound is lignin.
 - 12. The composition of claim 11, wherein said frequency is in the range of 4240-4260 MHz, and said field strength is in the range of 80-320 mV/cm.

13. The composition of claim 2, wherein said polymeric compound is polyethylene.

- 14. The composition of claim 13, wherein said frequency is in the range of 4240-4260 MHz, and said field strength is in the range of 60-270 mV/cm.
- 5 15. The composition of claim 2, wherein said polymeric compound is polypropylene.
 - 16. The composition of claim 15, wherein said frequency is in the range of 4240-4260 MHz, and said field strength is in the range of 90-320 mV/cm.
- 17. The composition of claim 2, wherein said polymeric compound is polyvinyl chloride.
 - 18. The composition of claim 17, wherein said frequency is in the range of 4240-4260 MHz, and said field strength is in the range of 70-350 mV/cm.
 - 19. The composition of claim 2, wherein said polymeric compound is polystyrene.
- 15 20. The composition of claim 19, wherein said frequency is in the range of 4240-4260 MHz, and said field strength is in the range of 60-260 mV/cm.
- 21. The composition of claim 1 comprising a plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to degrade a nitrogen-containing compound in a culture medium as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 5520 to 5540 MHz and a field strength in the range of 0.5 to 360 mV/cm, as compared to yeast cells not having been so cultured.

22. The composition of claim 21, wherein said frequency is in the range of 5521 to 5538 MHz.

23. The composition of claim 21, wherein said yeast cells are derived from cells of the species Saccharomyces cerevisiae or Saccharomyces carlsbergensis.

5

15

- 24. The composition of claim 21, wherein said yeast cells are derived from cells of the strain deposited at the China General Microbiological Culture Collection Center with an accession number selected from the group consisting of AS2.93, AS2.98, AS2.152, AS2.423, AS2.452, AS2.458, AS2.502, AS2.535, AS2.561, AS2.440 and AS2.595.
 - 25. The composition of claim 21, wherein said nitrogen-containing compound is animal protein.
 - 26. The composition of claim 25, wherein said frequency is in the range of 5521-5538 MHz and said field strength is in the range of 90-360 mV/cm.
 - 27. The composition of claim 21, wherein said nitrogen-containing compound is plant protein.
 - 28. The composition of claim 27, wherein said frequency is in the range of 5521-5538 MHz and said field strength is in the range of 120-340 mV/cm.
- 29. The composition of claim 1 comprising a plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to degrade an antibiotic or an organic solvent in a culture medium as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 70 to 100 MHz and a field strength in the range of 0.5 to 350 mV/cm, as compared to yeast cells not having been so cultured.

30. The composition of claim 29, wherein said yeast cells are derived from cells of the species Saccharomyces cerevisiae, Saccharomyces carlsbergensis, Saccharomyces rouxii, Saccharomyces willianus, or Candida utilis.

- 31. The composition of claim 29, wherein said yeast cells are derived
 from cells of the strain deposited at The China General Microbiological Culture
 Collection Center with an accession number selected from the group consisting of
 AS2.4, AS2.14, AS2.416, AS2.430, AS2.593, IFFI1002, IFFI1006, IFFI1043,
 IFFI1045, IFFI1048, IFFI1063, IFFI1059, IFFI1206, IFFI1209, IFFI1210,
 IFFI1211, IFFI1213, IFFI1215, IFFI1220, IFFI1221, IFFI1224, IFFI1248,
 IFFI1270, IFFI1290, IFFI1291, IFFI1293, IFFI1297, IFFI1301, IFFI1302,
 IFFI1310, IFFI1311, IFFI1331, IFFI1335, IFFI1336, IFFI1338, IFFI1339,
 IFFI1340, IFFI1345, IFFI1396, IFFI1399, IFFI1411, IFFI1413, AS2.293, AS2.377,
 AS2.444, AS2.178, and AS2.120.
 - 32. The composition of claim 29, wherein said antibiotic is penicillin.
- 15 33. The composition of claim 29, wherein said field strength is in the range of 30-220 mV/cm.
 - 34. The composition of claim 29, wherein said antibiotic is chlortetracycline.
- 35. The composition of claim 34, wherein said field strength is in the 20 range of 30-230 mV/cm.
 - 36. The composition of claim 29, wherein said antibiotic is oxytetracycline.
 - 37. The composition of claim 36, wherein said field strength is in the range of 30-250 mV/cm.

38. The composition of claim 29, wherein said antibiotic is doxycycline.

- 39. The composition of claim 38, wherein said field strength is in the range of 30-250 mV/cm.
 - 40. The composition of claim 29, wherein said antibiotic is tetracycline.
- 5 41. The composition of claim 40, wherein said field strength is in the range of 30-230 mV/cm.
 - 42. The composition of claim 29, wherein said antibiotic is streptomycin.
- 43. The composition of claim 42, wherein said field strength is in the 10 range of 30-230 mV/cm.
 - 44. The composition of claim 29, wherein said antibiotic is kanamycin.
 - 45. The composition of claim 44, wherein said field strength is in range of 30-230 mV/cm.
- 46. The composition of claim 29, wherein said antibiotic is erythromycin.
 - 47. The composition of claim 46, wherein said field strength is in the range of 30-250 mV/cm.
 - 48. The composition of claim 29, wherein said antibiotic is spiramycin.
- 49. The composition of claim 48, wherein said field strength is in the 20 range of 30-230 mV/cm.

50. The composition of claim 29, wherein said antibiotic is bacitracin.

- 51. The composition of claim 50, wherein said field strength is in the range of 30-230 mV/cm.
- 52. The composition of claim 29, wherein said organic solvent is trichloromethane.
 - 53. The composition of claim 52, wherein said field strength is in the range of 90-280 mV/cm.
 - 54. The composition of claim 29, wherein said organic solvent is toluene or ethylbenzene.
- The composition of claim 54, wherein said field strength is in the range of 80-280 mV/cm.
 - 56. The composition of claim 29, wherein said organic solvent is p-xylene.
- 57. The composition of claim 56, wherein said field strength is in the range of 80-280 mV/cm.
 - 58. The composition of claim 29, wherein said organic solvent is furazolidonum.
 - 59. The composition of claim 58, wherein said field strength is in the range of 80-280 mV/cm.
- 20 60. The composition of claim 29, wherein said organic solvent is decoquinate.

61. The composition of claim 60, wherein said field strength is in the range of 90-280 mV/cm.

- 62. The composition of claim 29, wherein said organic solvent is benzaldehyde.
- 5 63. The composition of claim 62, wherein said field strength is in the range of 120-280 mV/cm.
 - 64. The composition of claim 29, wherein said organic solvent is propylaldehyde.
- 65. The composition of claim 64, wherein said field strength is in the range of 100-200 mV/cm.
 - 66. The composition of claim 29, wherein said organic solvent is enanthaldehyde.
 - 67. The composition of claim 66, wherein said field strength is in the range of 110-280 mV/cm.
- 15 68. The composition of claim 29, wherein said organic solvent is m-dichlorobenzene.
 - 69. The composition of claim 68, wherein said field strength is in the range of 100-220 mV/cm.
- 70. The composition of claim 29, wherein said organic solvent is 20 acetophenone.
 - 71. The composition of claim 70, wherein said field strength is in the range of 116-225 mV/cm.

72. The composition of claim 29, wherein said organic solvent is arsanilic acid.

- 73. The composition of claim 72, wherein said field strength is in the range of 90-190 mV/cm.
- 5 74. The composition of claim 29, wherein said organic solvent is roxarsone.
 - 75. The composition of claim 74, wherein said field strength is in the range of 100-190 mV/cm.
- 76. The composition of claim 29, wherein said organic solvent is 10 dodecane.
 - 77. The composition of claim 76, wherein said field strength is in the range of 160-300 mV/cm.
 - 78. The composition of claim 29, wherein said organic solvent is nonadecane.
- 15 79. The composition of claim 78, wherein said field strength is in the range of 120-300 mV/cm.
 - 80. The composition of claim 29, wherein said organic solvent is octacosane.
- 81. The composition of claim 80, wherein said field strength is in the 20 range of 120-300 mV/cm.

82. The composition of claim 29, wherein said organic solvent is trichlorphonum.

- 83. The composition of claim 82, wherein said field strength is in the range of 200-300 mV/cm.
- 5 84. The composition of claim 29, wherein said organic solvent is dinitornidum or zoalene.
 - 85. The composition of claim 84, wherein said field strength is in the range of 130-310 mV/cm.
- 86. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to degrade trichloromethane in a culture medium as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 126 to 142 MHz and a field strength in the range of 90 to 280 mV/cm, as compared to yeast cells not having been so cultured.
- 15 87. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to degrade toluene or ethylbenzene in a culture medium as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 52 to 70 MHz and a field strength in the range of 80 to 280 mV/cm, as compared to yeast cells not having been so cultured.
 - 88. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to degrade p-xylene in a culture medium as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 30 to 50 MHz and a field strength in the range of 80 to 280 mV/cm, as compared to yeast cells not having been so cultured.

89. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to degrade furazolidonum in a culture medium as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 200 to 220 MHz and a field strength in the range of 80 to 280 mV/cm, as compared to yeast cells not having been so cultured.

- 90. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to degrade decoquinate in a culture medium as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 213 to 229 MHz and a field strength in the range of 90 to 280 mV/cm, as compared to yeast cells not having been so cultured.
- 91. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to degrade benzaldehyde in a culture medium as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 133 to 151 MHz and a field strength in the range of 120 to 280 mV/cm, as compared to yeast cells not having been so cultured.
- 92. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to degrade propylaldehyde in a culture medium as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 145 to 162 MHz and a field strength in the range of 100 to 200 mV/cm, as compared to yeast cells not having been so cultured.
- 93. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to degrade enanthaldehyde in a culture medium as a result of having been cultured

in the presence of an alternating electric field having a frequency in the range of 156 to 176 MHz and a field strength in the range of 110 to 280 mV/cm, as compared to yeast cells not having been so cultured.

- 94. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to degrade m-dichlorobenzene in a culture medium as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 163 to 183 MHz and a field strength in the range of 100 to 220 mV/cm, as compared to yeast cells not having been so cultured.
- 10 95. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to degrade acetophenone in a culture medium as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 175 to 191 MHz and a field strength in the range of 116 to 225 mV/cm, as compared to yeast cells not having been so cultured.
- 96. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to degrade arsanilic acid in a culture medium as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 183 to 205 MHz and a field strength in the range of 90 to 190 mV/cm, as compared to yeast cells not having been so cultured.
 - 97. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to degrade roxarsone in a culture medium as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 114 to 128 MHz and a field strength in the range of 100 to 190 mV/cm, as compared to yeast cells not having been so cultured.

25

98. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to degrade dodecane in a culture medium as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 244 to 264 MHz and a field strength in the range of 160 to 300 mV/cm, as compared to yeast cells not having been so cultured.

- 99. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to degrade nonadecane or octacosane in a culture medium as a result of having
 10 been cultured in the presence of an alternating electric field having a frequency in the range of 252 to 278 MHz and a field strength in the range of 120 to 300 mV/cm, as compared to yeast cells not having been so cultured.
 - 100. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to degrade trichlorophonum in a culture medium as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 220 to 250 MHz and a field strength in the range of 200 to 300 mV/cm, as compared to yeast cells not having been so cultured.
- plurality of yeast cells are characterized by a substantial increase in their capability to degrade dinitomidum or zoalene in a culture medium as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 220 to 250 MHz and a field strength in the range of 130 to 310 mV/cm, as compared to yeast cells not having been so cultured.
- 25 102. The composition of claim 1 comprising a plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to convert bio-available nitrogen in a culture medium into intracellular nitrogen as a result of having been cultured in the presence of an

alternating electric field having a frequency in the range of 660 to 680 MHz and a field strength in the range of 0.1 to 350 mV/cm, as compared to yeast cells not having been so cultured.

- 103. The composition of claim 102, wherein said field strength is in the range of 100 to 350 mV/cm.
 - 104. The composition of claim 102, wherein said yeast cells are derived from cells of the species Saccharomyces cerevisiae, Saccharomyces willianus, Candida tropicalis or Geotrichum candidum.
- 105. The composition of claim 102, wherein said yeast cells are derived from cells of the strain deposited at The China General Microbiological Culture Collection Center with an accession number selected from the group consisting of AS2.152, AS2.196, AS2.336, AS2.400, AS2.416, AS2.423, AS2.498, AS2.614, AS2.982, and AS2.1387.
- 106. The composition of claim 102, wherein the bio-available nitrogen is ammonium.
 - 107. The composition of claim 106, wherein said frequency is in the range of 660 to 680 MHz and said field strength is in the range of 140-320 mV/cm.
 - 108. The composition of claim 102, wherein the bio-available nitrogen is nitrate or nitrite.
- 20 109. The composition of claim 108, wherein said frequency is in the range of 660 to 680 MHz and said field strength is in the range of 120-290 mV/cm.
 - 110. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to convert ammonium in a culture medium into intracellular nitrogen as a result of

having been cultured in the presence of an alternating electric field having a frequency in the range of 2160 to 2190 MHz and a field strength in the range of 140 to 320 mV/cm, as compared to yeast cells not having been so cultured.

- 111. The composition of claim 1 comprising a plurality of yeast cells,
 5 wherein said plurality of yeast cells are characterized by a substantial increase in their capability to convert biologically available phosphorus in a culture medium into intracellular phosphorus as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 80 to 440 MHz and a field strength in the range of 0.5 to 350 mV/cm, as compared to yeast cells not
 10 having been so cultured.
 - 112. The composition of claim 111, wherein said frequency is in the range of 86 to 120 MHz or 410 to 430 MHz.
- 113. The composition of claim 111, wherein said yeast cells are derived from cells of the species Saccharomyces cerevisiae or Saccharomyces
 15 carlsbergensis.
 - 114. The composition of claim 111, wherein said yeast cells are derived from cells of the strain deposited at The China General Microbiological Culture Collection Center with an accession number selected from the group consisting of AS2.346, AS2.423, AS2.430, AS2.451, AS2.558, AS2.620, AS2.628, IFFI 1203, and AS2.189.
 - 115. The composition of claim 111, wherein said biologically available phosphorus is PO₄³⁻, HPO₄²⁻, H₂PO₄, or H₃PO₄.

20

116. The composition of claim 115, wherein said frequency is in the range of 86 to 120 MHz and said field strength is in the range of 60 to 260 mV/cm.

117. The composition of claim 1 comprising a plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to reduce odor of a culture medium as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 2160 to 2380 MHz and a field strength in the range of 0.5 to 320 mV/cm, as compared to yeast cells not having been so cultured.

- 118. The composition of claim 117, wherein said frequency is in the range of 2160 to 2250 MHz or 2280 to 2380 MHz.
- 119. The composition of claim 117, wherein said yeast cells are derived from cells of the species Saccharomyces cerevisiae or Saccharomyces carlsbergensis.
 - 120. The composition of claim 117, wherein said yeast cells are derived from cells of the strain deposited at The China General Microbiological Culture Collection Center with an accession number selected from the group consisting of AS2.53, AS2.163, AS2.396, AS2.397, AS2.423, AS2.452, AS2.502, AS2.516, AS2.541, AS2.558, AS2.559, AS2.560, AS2.561, AS2.562, AS2.605, AS2.607, AS2.612, IFFI 1052, IFFI 1202, IFFI 1213, IFFI 1247, and IFFI 1397.
 - 121. The composition of claim 117, wherein said odor is caused by hydrogen sulfide.
- 20 122. The composition of claim 121, wherein said frequency is in the range of 2160 to 2250 MHz, and said field strength is in the range of 80 to 250 mV/cm.
 - 123. The composition of claim 117, wherein said odor is caused by ammonia.

124. The composition of claim 123, wherein said frequency is in the range of 2160 to 2250 MHz, and said field strength is in the range of 70 to 260 mV/cm.

- 125. The composition of claim 117, wherein said odor is caused by 5 indole.
 - 126. The composition of claim 125, wherein said frequency is in the range of 2160 to 2250 MHz, and said field strength is in the range of 70 to 260 mV/cm.
- 127. The composition of claim 117, wherein said odor is caused by methylamine, dimethylamine, or trimethylamine.
 - 128. The composition of claim 127, wherein said frequency is in the range of 2160 to 2250 MHz, and said field strength is in the range of 40 to 260 mV/cm.
- 129. The composition of claim 117, wherein said odor is caused by organic acids.
 - 130. The composition of claim 129, wherein said frequency is in the range of 2280 to 2380 MHz, and said field strength is in the range of 140 to 300 mV/cm.
- 131. The composition of claim 130, wherein said odor is caused by acetic 20 acid.
 - 132. The composition of claim 117, wherein said odor is caused by p-cresol.

133. The composition of claim 132, wherein said frequency is in the range of 2280 to 2380 MHz, and said field strength is in the range of 90 to 260 mV/cm.

- 134. The composition of claim 1 comprising a plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to suppress the growth of pathogenic microbes as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 30 to 50 MHz and a field strength in the range of 0.5 to 200 mV/cm, as compared to yeast cells not having been so cultured.
- 10 135. The composition of claim 134, wherein said field strength is in the range of 10 to 180 mV/cm.
 - 136. The composition of claim 134, wherein said yeast cells are derived from cells of the species Saccharomyces cerevisiae, Saccharomyces carlsbergensis, Saccharomyces uvarum, or Saccharomyces willianus.
- 137. The composition of claim 134, wherein said yeast cells are derived from cells of the strain deposited at The China General Microbiological Culture Collection Center with an accession number selected from the group consisting of ACCC2034, ACCC2043, AS2.70, AS2.119, AS2.152, AS2.200, AS2.369, AS2.408, AS2.451, AS2.562, AS2.607, IFFI1021, IFFI1023, IFFI1032, IFFI1037, IFFI1205, IFFI1211, IFFI1221, IFFI1251, IFFI1301, IFFI1307, IFFI1308, IFFI1331, and IFFI1345.
 - 138. The composition of claim 134, wherein said pathogenic microbe is Staphylococcus aures.
- 139. The composition of claim 134, wherein said pathogenic microbe is 25 Diplococcus pneumonia.

140. The composition of claim 134, wherein said pathogenic microbe is *Bacillus anthracis*.

- 141. The composition of claim 134, wherein said pathogenic microbe is Mycobacterium tuberculosis.
- 5 142. The composition of claim 134, wherein said pathogenic microbe is *E. Coli*.
 - 143. The composition of claim 134, wherein said pathogenic microbe is Salmonella.
- 144. The composition of claim 1 comprising a plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to suppress the growth of algae as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 6340 to 6380 MHz and a field strength in the range of 0.5 to 400 mV/cm, as compared to yeast cells not having been so cultured.
- 15 The composition of claim 144, wherein said frequency is in the range of 6352 to 6370 MHz.
 - 146. The composition of claim 144, wherein said field strength is in the range of 60 to 380 mV/cm.
- 147. The composition of claim 144, wherein said yeast cells are derived 20 from cells of the species Saccharomyces cerevisiae.
 - 148. The composition of claim 144, wherein said yeast cells are derived from cells of the strain deposited at the China General Microbiological Culture Collection Center with an accession number selected from the group consisting of

AS2.408, AS2.414, AS2.416, AS2.422, AS2.453, AS2.486, AS2.558, AS2.562, and IFFI1292.

- 149. The composition of claim 145, wherein said algae are green algae.
- 150. The composition of claim 149, wherein said field strength is in the range of 100 to 330 mV/cm.
 - 151. The composition of claim 145, wherein said algae are blue algae.
 - 152. The composition of claim 151, wherein said field strength is in the range of 70 to 310 mV/cm.
 - 153. The composition of claim 145, wherein said algae are red algae.
- 10 154. The composition of claim 153, wherein said field strength is in the range of 120 to 360 mV/cm.
 - 155. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells are characterized by a substantial increase in their capability to decompose debris of algae as a result of having been cultured in the presence of an alternating electric field having a frequency in the range of 4440 to 4470 MHz and a field strength in the range of 0.5 to 400 mV/cm, as compared to yeast cells not having been so cultured.
 - 156. The composition of claim 155, wherein said algae are green, blue or red algae.
- 20 157. The composition of claim 156, wherein said frequency is in the range of 4452 to 4470 MHz and said field strength is in the range of 50 to 280 mV/cm.

158. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells have been activated such that they have a substantially increased capability to degrade a polymeric compound in a culture medium as compared to unactivated yeast cells.

- 159. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells have been activated such that they have a substantially increased capability to degrade a nitrogen-containing compound in a culture medium as compared to unactivated yeast cells.
- 160. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells have been activated such that they have a substantially increased capability to degrade an antibiotic or an organic solvent in a culture medium as compared to unactivated yeast cells.
- 161. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells have been activated such that they have a substantially
 15 increased capability to convert bio-available nitrogen in a culture medium into intracellular nitrogen as compared to unactivated yeast cells.
 - 162. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells have been activated such that they have a substantially increased capability to convert biologically available phosphorus in a culture medium into intracellular phosphorus as compared to unactivated yeast cells.

20

- 163. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells have been activated such that they have a substantially increased capability to reduce odor of a culture medium as compared to unactivated yeast cells.
- 25 164. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells have been activated such that they have a substantially

increased capability to suppress the growth of pathogenic microbes as compared to unactivated yeast cells.

- 165. A composition comprising a plurality of yeast cells, wherein said plurality of yeast cells have been activated such that they have a substantially increased capability to suppress the growth of algae or decompose debris of algae as compared to unactivated yeast cells.
- plurality of yeast cells in the presence of an alternating electric field having a frequency in the range of 4230 to 4260 MHz and a field strength in the range of 0.5 to 360 mV/cm, wherein resulting plurality of yeast cells are characterized by a substantial increase in their capability to degrade a polymeric compound in a culture medium as a result of said culturing as compared to yeast cells not having been so cultured.
- 167. A method of preparing a yeast composition, comprising culturing a plurality of yeast cells in the presence of an alternating electric field having a frequency in the range of 5520 to 5540 MHz and a field strength in the range of 0.5 to 360 mV/cm, wherein resulting plurality of yeast cells are characterized by a substantial increase in their capability to degrade a nitrogen-containing compound in a culture medium as a result of said culturing as compared to yeast cells not having been so cultured.
 - 168. A method of preparing a yeast composition, comprising culturing a plurality of yeast cells in the presence of an alternating electric field having a frequency in the range of 70 to 100 MHz and a field strength in the range of 0.5 to 350 mV/cm, wherein resulting plurality of yeast cells are characterized by a substantial increase in their capability to degrade an antibiotic or an organic solvent in a culture medium as a result of said culturing as compared to yeast cells not having been so cultured.

25

169. A method of preparing a yeast composition, comprising culturing a plurality of yeast cells in the presence of an alternating electric field having a frequency in the range of 660 to 680 MHz and a field strength in the range of 0.1 to 350 mV/cm, wherein resulting plurality of yeast cells are characterized by a substantial increase in their capability to convert bio-available nitrogen in a culture medium into intracellular nitrogen as a result of said culturing as compared to yeast cells not having been so cultured.

- 170. A method of preparing a yeast composition, comprising culturing a plurality of yeast cells in the presence of an alternating electric field having a frequency in the range of 80 to 440 MHz and a field strength in the range of 0.5 to 350 mV/cm, wherein resulting plurality of yeast cells are characterized by a substantial increase in their capability to convert biologically available phosphorus in a culture medium into intracellular phosphorus as a result of said culturing as compared to yeast cells not having been so cultured.
- 15 171. A method of preparing a yeast composition, comprising culturing a plurality of yeast cells in the presence of an alternating electric field or multiple alternating electric fields having a frequency in the range of 2160 to 2380 MHz and a field strength in the range of 0.5 to 320 mV/cm, wherein resulting plurality of yeast cells are characterized by a substantial increase in their capability to reduce odor of a culture medium as a result of said culturing as compared to yeast cells not having been so cultured.
- 172. A method of preparing a yeast composition, comprising culturing a plurality of yeast cells in the presence of an alternating electric field having a frequency in the range of 30 to 50 MHz and a field strength in the range of 0.5 to
 25 200 mV/cm, wherein resulting plurality of yeast cells are characterized by a substantial increase in their capability to suppress the growth of pathogenic microbes as a result of said culturing as compared to yeast cells not having been so cultured.

173. A method of preparing a yeast composition, comprising culturing a plurality of yeast cells in the presence of an alternating electric field having a frequency in the range of 6340 to 6380 MHz and a field strength in the range of 0.5 to 400 mV/cm, wherein resulting plurality of yeast cells are characterized by a substantial increase in their capability to suppress the growth of algae as a result of said culturing as compared to yeast cells not having been so cultured.

- 174. A method of preparing a yeast composition, comprising culturing a plurality of yeast cells in the presence of an alternating electric field having a frequency in the range of 4440 to 4470 MHz and a field strength in the range of 0.5 to 400 mV/cm, wherein resulting plurality of yeast cells are characterized by a substantial increase in their capability to decompose debris of algae as a result of said culturing as compared to yeast cells not having been so cultured.
- 175. A method of treating waste, comprising the step of administering one or more compositions of any one of claims 1, 21, 29, 86-102, 110, 111, 117, 134, 144, 155, or 158-165 to said waste.
 - 176. The method of claim 175, wherein said waste is waste water.
 - 177. The method of claim 176, wherein said waste water is sewage.
 - 178. The method of claim 176, wherein said waste water is industrial waste water.
- 20 179. The method of claim 176, wherein said waste water is surface water.
 - 180. The method of claim 176, wherein said waste water is drinking water.

This Page is Inserted by IFW Indexing and Scanning Operations and is not part of the Official Record

BEST AVAILABLE IMAGES

Defective images within this document are accurate representations of the original documents submitted by the applicant.

Defects in the images include but are not limited to the items checked:

BLACK BORDERS

IMAGE CUT OFF AT TOP, BOTTOM OR SIDES

FADED TEXT OR DRAWING

BLURRED OR ILLEGIBLE TEXT OR DRAWING

SKEWED/SLANTED IMAGES

COLOR OR BLACK AND WHITE PHOTOGRAPHS

GRAY SCALE DOCUMENTS

LINES OR MARKS ON ORIGINAL DOCUMENT

REFERENCE(S) OR EXHIBIT(S) SUBMITTED ARE POOR QUALITY

IMAGES ARE BEST AVAILABLE COPY.

☐ OTHER:

As rescanning these documents will not correct the image problems checked, please do not report these problems to the IFW Image Problem Mailbox.

THIS PAGE BLANK (USPTO)